Factors associated with secondary immune thrombocytopenia in patients with primary Sjögren's syndrome: a retrospective study of 639 cases

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Abstract

Objective

To investigate the clinical characteristics and relevant factors of secondary immune thrombocytopenia (ITP) in patients with primary Sjögren's syndrome (pSS).

Methods

Patients with pSS being treated between 2013 and 2020 in China-Japan Friendship Hospital were retrospectively analysed. Clinical characteristics were compared between pSS patients with and without secondary ITP. Logistic regression analysis was performed to identify factors associated with secondary ITP in patients with pSS.

Results

639 patients with pSS were included in this study, among which 566 (88.6%) were women. The prevalence of secondary ITP in patients with pSS were 12.4%. Among pSS patients with secondary ITP, 55.7% had mucocutaneous bleeding and 8.9% experienced visceral bleeding. Lymphopenia (OR=3.154, 95% CI 1.185-8.395, p=0.021), anaemia (OR=2.416, 95% CI 1.250-4.668, p=0.009), low C4 (OR=2.904, 95% CI 1.563-5.394, p=0.001), and positive anti-RNP (OR=2.777, 95% CI 1.070-7.202, p=0.036) were significantly related to secondary ITP, while interstitial lung disease (ILD, OR=0.429, 95% CI 0.203-0.907, p=0.027), ANA ≥1:320 (OR=0.469, 95% CI 0.221-0.996, p=0.049) and positive anti-SSB (OR=0.288, 95% CI 0.126-0.685, p=0.003) were negatively associated with secondary ITP in patients with pSS.

Conclusion

Over 10% of patients with pSS had secondary ITP, among whom visceral bleeding was comparatively rare. Lymphopenia and anaemia were positively related to secondary ITP, while ILD was negatively associated with secondary ITP. Low C4 and positive anti-RNP seem to be two potential risk factors for secondary ITP in patients with pSS, while ANA ≥1:320 and positive anti-SSB may be two potential protective factors.

Key words

Sjögren's syndrome, immune thrombocytopenia, platelet, risk factor, clinical characteristic

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Introduction

Primary Sjögren's syndrome (pSS) is a common systemic autoimmune disease, characterised by B-cell hyperactivity and lymphocytic infiltration of exocrine glands and extra-glandular organs (1, 2). Dryness of mouth and eyes are the most prevalent symptoms of this disease, while more than 30% of the patients may present systemic manifestations ranging from fatigue, arthralgia, interstitial lung disease (ILD), thrombocytopenia or hypokalaemia (1, 3). These symptoms and organ involvement affect health of the patients and their quality of life (4, 5). With a female-to-male ratio of 9:1 and peak incidence at approximately 50 years of age, pSS affects 1 to 23 persons per 10 000 inhabitants in European countries, 2 to 10 per 10000 inhabitants in US population, and in approximately 6 per 10000 Chinese inhabitants (6-8).

Immune thrombocytopenia (ITP) is an acquired thrombocytopenia, defined as a platelet count below 100×10^9/L, with other causes of thrombocytopenia ruled out (9). ITP can be further differentiated into primary ITP and secondary ITP. The latter is commonly associated with autoimmune diseases, such as pSS and systematic lupus erythematosus (SLE) (10, 11). Since thrombocytopenia was associated with metrics of the disease's activity and prognosis in pSS (12), exploring factors associated with secondary ITP in patients with pSS can provide insights in clinical management of this disease. Currently, a few studies have investigated the clinical characteristics of ITP in patients with pSS. A Korean cohort study (13) included 113 pSS patients showed that 23.9% patients with pSS had autoimmune cytopenia, but no further analysis of thrombocytopenia was conducted. A Chinese retrospective study evaluated features of ITP associated with autoimmune diseases reported that 18.82% pSS patients complicated with thrombocytopenia (8). Another recent single-centre retrospective study (14) enrolling 291 Chinese pSS patients analysed the clinical characteristics of ITP in patients with pSS and reported that 12.03% patients with pSS had secondary ITP. However, characteristics of ITP associated with pSS were not understood fully and it remains unclear about factors associated with ITP in patients with pSS in previous studies due to the limited sample size. Therefore, we conducted this retrospective study to further investigate clinical characteristics and relevant factors of secondary ITP in patients with pSS to inform clinical work.

Materials and methods

Study population

Outpatients and inpatients with pSS being treated in China-Japan Friendship Hospital between January 2013 and December 2020 were retrospectively analysed. The diagnosis of pSS was based on the 2002 American-European Consensus Group criteria for pSS (15) or the 2016 American College of Rheumatology/European League Against Rheumatism classification criteria (16). ITP was diagnosed according to the American Society of Haematology guidelines (17) and all ITP included in this study were associated with pSS. All pSS patients with secondary ITP had a platelet count below 100×10^9/L. Severe ITP is defined as peripheral blood platelet count less than 20×10^9/L (18). Patients with cancer or secondary SS were excluded. Patients developed thrombocytopenia due to other reasons such as primary haematological disease, liver cirrhosis, antiphospholipid syndrome, or infection were excluded. Pregnant women were excluded. If a record missed important data that was used to diagnose pSS, or platelet count, the record would also be excluded. This study was approved by the Clinical Research Ethics Committee of China-Japan Friendship Hospital (no. 2021-144-K102). Informed consent was exempted because the datasets were devoid of personally identifiable information.

Date collection

Clinical data including patient demographics, clinical manifestations, laboratory indicators, and diagnosis were obtained from medical records of eligible patients and the hospital information system. If a patient was treated more than one time, only medical record of the first encounter associated with pSS was included for analysis. All manifes-

tations were either concomitant or before the thrombopenia diagnosis. Clinical manifestations included symptoms related to pSS such as dryness, fatigue, and arthralgia. ILD was detected by high-resolution computed tomography and evaluated by two experienced radiologists. Abnormal Schirmer I test was defined as a result ≤5mm/5min. Laboratory indicators included full blood test, immunoglobulin A (IgA), immunoglobulin G (IgG), immunoglobulin M (IgM), complement 3 (C3), and complement 4 (C4), erythrocyte sedimentation rate (ESR), antinuclear antibodies (ANA), rheumatoid factor (RF), anti-Sjögren's syndrome A (SSA) antibody, anti-Ro52 antibody, anti-Sjögren's syndrome B (SSB) antibody, anti-centromere protein B (CENP-B) antibody, anti-ribonucleoprotein (RNP) antibody, anti-cardiolipin (ACL) and anti-β2-glycoprotein I (β2GPI) antibodies.

All tests were performed using commercial techniques standardised in China-Japan Friendship Hospital. Leucopenia was defined as white blood cell count <4.00 x 10^9/L, neutropenia was defined as neutrophil <1.5 × 10^9/L, anaemia was defined as haemoglobin concentration <110 g/L, and thrombocytopenia was defined as platelet count <100 × 10^9/L. Hypergammaglobulinaemia was defined as IgA >3.78 g/L, IgG >16.2 g/L, or IgM >2.63 g/L. Hypocomplementaemia was defined as C3 <0.7 g/L, or low C4 <0.16 g/L. Elevated ESR was defined as ESR >20 mm/h. The ANA titre was determined by an indirect immunofluorescence assay on HEp2 cells, and a titre of 1:320 was considered positive. RF was determined by immunoturbidimetric assay, and positivity defined as a level over 20 IU/mL. Anti-ACL and anti-β2GPI antibodies were tested using commercial enzyme-linked immunosorbent assay kits. Anti-SSA and all other autoantibodies were tested using commercial immunoblot kits. Positive minor salivary gland (MSG) biopsy was defined as focal lymphocytic sialadenitis with a focus score of ≥1 focus per 4 mm^2 (15).

Statistical analysis

SPSS (v. 19.0) software was used for data analyses. Continuous data were presented as median with interquartile range (IQR) for any non-normally distribution. Categorical data were presented as numbers and percentages. The Shapiro-Wilk test was conducted to detect whether the data were normally distributed. In the between-group comparisons, the Mann-Whitney U-test was used for continuous data which did not normally distributed, and the Chi-squared test or Fisher's exact test as appropriate was conducted to compare binary data. Multivariate logistic regression analysis was performed to identify factors associated with ITP in patients with pSS, adjusted for age (the age of patients when they visited the China-Japan Friendship Hospital due to pSS), sex, disease duration (time from onset to diagnosis of pSS in China-Japan Friendship Hospital) and those with statistical *p*-value <0.1 in univariate analyses. The results were presented as odds ratio (OR) with its 95% confidence interval (CI). A two-sided *p*-value <0.05 was considered significant.

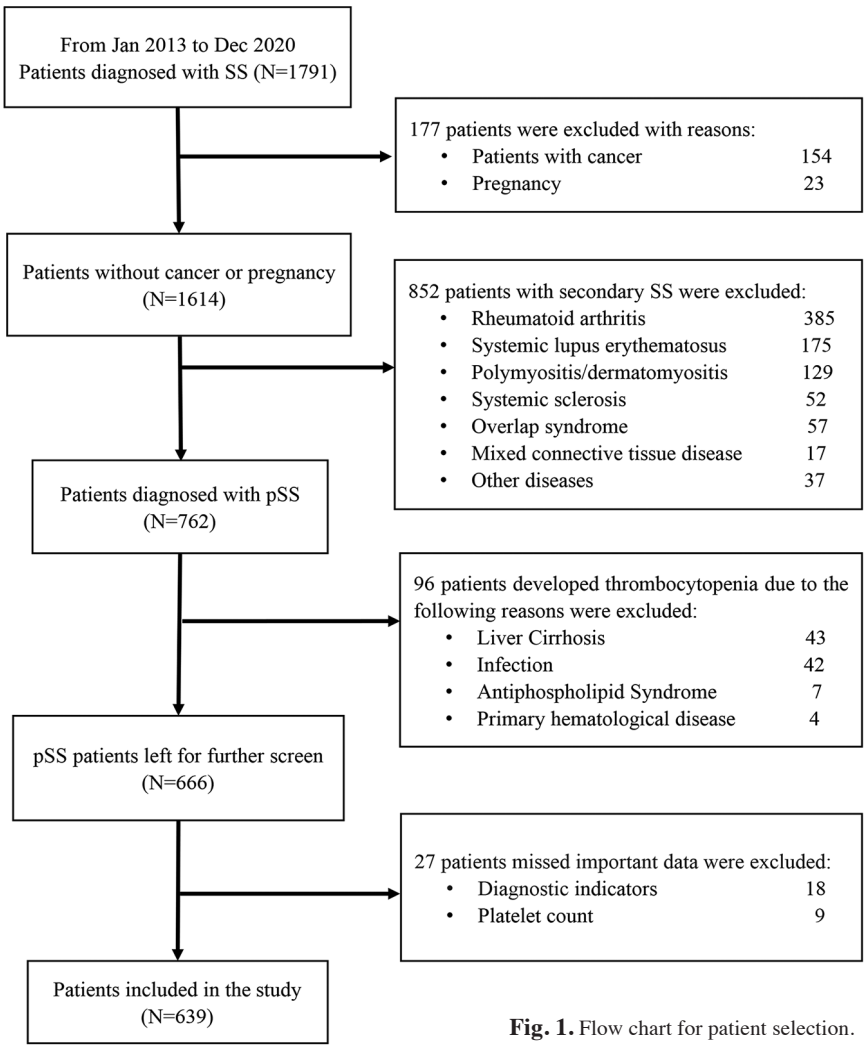

Fig. 1. Flow chart for patient selection.

Results

Demographic data of pSS patients with and without secondary ITP

A total of 639 Chinese patients with pSS, involving 79 (12.4%) with secondary ITP and 560 (87.6%) without ITP, were included in this study (Fig. 1). 566 (88.6%) were women, with a median age of 57 years (IQR 48-64) years old. The median duration of pSS was 36 months (IQR 10-96). There was no obvious difference in age, gender, and disease duration between pSS patients with and without secondary ITP (Table I).

Clinical manifestations of pSS patients with and without secondary ITP

As shown in Table I, patients with pSS

commonly had dry mouth (85.9%) and dry eyes (79.7%). Majority of (92.0%) patients were consistent with Schirmer I test≤5 mm/5 min in ocular evaluation. Some patients had fatigue (46.2%), arthralgia (41.8%), dental caries (39.7%), or parotid enlargement (17.1%). ILD was found in 225 patients with pSS (35.2%). 369 patients (57.7%) included in this study achieved MSG biopsy, and 98.6% of the 369 patients presented positive findings, with a median MSG focus score of 3 (IQR 1-4). The clinical manifestations including dry mouth, dry eyes, Schirmer I test ≤5 mm/5 min, fatigue, arthralgia, dental caries, parotid enlargement, positive MSG biopsy, and MSG focus score were comparable between pSS patients with and without secondary ITP. However, pSS patients with secondary ITP had lower prevalence of ILD, compared with pSS patients without ITP (22.8% *vs*. 37.0%, p=0.014).

Table I. Clinical characteristics of pSS patients with and without secondary ITP.

Characteristics	Total (n=639)	pSS with ITP (n=79)	pSS without ITP (n=560)	p-value**
Age (years)	57.0 (48.0-64.0)	58.0 (49.0-66.0)	56.0 (48.0-64.0)	0.252
Gender (female)	566 (88.6)	74 (93.7)	492 (87.9)	0.128
Disease duration (months)	36.0 (10.0-96.0)	48.0 (12.0-96.0)	36.0 (9.25-96.0)	0.539
Dry mouth	549 (85.9)	65 (82.3)	484 (86.4)	0.321
Dry eyes	509 (79.7)	61 (77.2)	448 (80.0)	0.565
Schirmer I test ≤ 5 mm/5 min	588 (92.0)	71 (89.9)	517 (92.3)	0.452
Fatigue	295 (46.2)	39 (49.4)	256 (45.7)	0.542
Arthralgia	264/631 (41.8)	26/75 (34.7)	238/556 (42.8)	0.180
Dental caires	254 (39.7)	36 (45.6)	218 (38.9)	0.259
Parotid enlargement	109 (17.1)	10 (12.7)	99 (17.7)	0.267
Interstitial lung disease	225 (35.2)	18 (22.8)	207 (37.0)	0.014
Minor salivary gland biopsy positivity*	364/369 (98.6)	32/32 (100.0)	332/337 (98.5)	1.000***
Focus score	3 (1-4)	2 (1-3)	3 (1.5-4)	0.200

All values are presented as n (%) or median (interquartile). ITP: immune thrombocytopenia.
*Positive minor salivary gland biopsy is defined as focal lymphocytic sialadenitis with a focus score of ≥1 focus/4 mm^2.
**Mann-Whitney U-test for continuous data, Chi-squared test for compare binary data.
***Fisher's exact test.

Platelets and bleeding of pSS patients with secondary ITP

Among the 79 patients (12.4%) who were complicated with ITP associated with pSS, 32 (40.5% of 79, 5.0% of 639) had severe ITP. The median level of platelets in pSS patients with secondary ITP was 29.0 (8.0-69.0) × 10^9/L, which is significantly lower than the median level of 206.0 (169.0-250.0) × 10^9/L in pSS patients without ITP (p<0.001) (Table II). The median level of platelets in pSS patients with severe ITP was 7.0 (2.0-12.8) × 10^9/L.

Compared with pSS patients without ITP, patients with secondary ITP had significantly higher proportion in mucocutaneous bleeding (55.7% *vs*. 3.0%, p<0.001) and visceral bleeding (8.9% *vs*. 0.2%, p<0.001). Visceral bleeding in pSS patients with secondary ITP included gastrointestinal bleeding (2.5%, n=2), gastrointestinal bleeding combined with haematuria (1.3%, n=1), vaginal bleeding (2.5%, n=2) and intracranial haemorrhage (2.5%, n=2); while in patients without ITP just contained gastrointestinal bleeding (0.2%, n=1) (Fig. 2).

Among pSS patients with secondary ITP, patients with intracranial hemorrhage all accompanied by severe ITP (platelet ≤3 × 10^9/L), without other causes of intracranial haemorrhage such as hypertension and brain arteriovenous malformation. Compared with pSS patients with non-severe ITP (platelet count ≥20 × 10^9/L and <100 × 10^9/L), patients with severe ITP (platelet count <20 × 10^9/L) had higher incidence in mucocutaneous bleeding (96.9% *vs*. 36.2%, p<0.001), but similar incidence in visceral bleeding (12.5% *vs*. 6.4%, p=0.592).

Haematological and serological characteristics of pSS patients with and without secondary ITP

Significant differences were also found in many laboratory indicators of pSS patients with and without secondary ITP (Table II). Besides platelet, pSS patients with secondary ITP had lower level of haemoglobin [116.0 (104.0-131.0) g/L *vs*. 125.0 (115.0-134.0) g/L, p<0.001], compared with pSS patients without ITP. Additionally, compared with patients without ITP, the proportion of lymphopenia was higher in patients with secondary ITP (16.5% *vs*. 5.9%, p=0.001), and similar finding with anaemia (31.6% *vs*. 15.7%, p=0.001). The differences between pSS patients with and without secondary ITP in other laboratory findings including leukocyte count, neutrophile count, lymphocyte count, the proportion of leucopenia, and the proportion of neutropenia were not significant.

Compared with pSS patients without ITP, pSS patients with secondary ITP had significantly lower levels of C3 [0.78 (0.66-0.90) g/L *vs*. 0.85 (0.74-0.97) g/L, p=0.002] and C4 [0.15 (0.11-0.19) g/L *vs*. 0.18 (0.15-0.22) g/L, p<0.001]. Furthermore, pSS patients with secondary ITP showed higher proportions of low C3 (33.3% *vs*. 19.6%, p=0.006), and low C4 (55.1% *vs*. 31.7%, p<0.001). No significant differences were detected between the two groups in immunoglobulin level and ESR.

As shown in Table II about the immunological characteristics, ANA titres ≥ 1:320 were found in 30.0% of included patients, and positive RF, anti-SSA, anti-Ro52, anti-SSB, anti-CENP-B, anti-RNP, anti-ACL, and anti-β2GPI were in 45.8%, 65.9%, 54.8%, 27.5%, 7.6%, 8.4%, 5.3%, and 5.3% of included patients, respectively. Compared with pSS patients without ITP, patients with secondary ITP had lower positivity of anti-SSB (16.7% *vs*. 29.1%, p=0.022). No significant differences were detected between the two groups in other autoantibodies.

Factors associated with secondary ITP in pSS patients

Logistic regression analysis was performed to identify factors that might be used to predict the development of secondary ITP in patients with pSS (Table III). Univariate analysis was performed

Table II. Haematological and serological characteristics of pSS patients with and without secondary ITP.

Variables	Total (n=639)	pSS with ITP (n=79)	pSS without ITP (n=560)	*p*-value*
Leukocyte ($\times10^9$/L)	5.04 (3.92-6.64)	4.87 (3.91-7.40)	5.05 (3.92-6.55)	0.683
Leucopenia (<4×10^9/L)	168 (26.3)	21 (26.6)	147 (26.3)	0.950
Neutrophile ($\times10^9$/L)	2.93 (2.12-4.17)	3.48 (2.17-4.64)	2.89 (2.11-4.09)	0.104
Neutropenia (<1.5×10^9/L)	44 (6.9)	7 (8.9)	37 (6.6)	0.459
Lymphocyte ($\times10^9$/L)	1.49 (1.13-1.85)	1.34 (0.97-1.87)	1.51 (1.15-1.85)	0.062
Lymphopenia (<0.8×10^9/L)	46 (7.2)	13 (16.5)	33 (5.9)	0.001
Haemoglobin (g/L)	125.0 (114.0-134.0)	116.0 (104.0-131.0)	125.0 (115.0-134.0)	<0.001
Anaemia (haemoglobin <110 g/L)	113 (17.7)	25 (31.6)	88 (15.7)	0.001
Platelet ($\times10^9$/L)	195.0 (153.0-241.0)	29.0 (8.0-69.0)	206 (169.0-250.0)	<0.001
Immunoglobulin A (g/L)	2.76 (1.91-3.77)	2.49 (1.90-3.52)	2.79 (1.91-3.81)	0.209
Hyper-immunoglobulin A (>3.78 g/L)	154/626 (24.6)	14/78 (17.9)	140/548 (25.5)	0.145
Immunoglobulin G (g/L)	15.50 (12.40-20.50)	15.35 (12.15-22.10)	15.50 (12.50-20.28)	0.804
Hyper-immunoglobulin G (>16.2 g/L)	281/626 (44.9)	36/78 (46.2)	245/548 (44.7)	0.810
Immunoglobulin M (g/L)	1.07 (0.74-1.58)	1.19 (0.77-1.94)	1.07 (0.73-1.55)	0.084
Hyper-immunoglobulin M (>2.63 g/L)	48/626 (7.7)	8/78 (10.3)	40/548 (7.3)	0.358
C3 (g/L)	0.84 (0.72-0.96)	0.78 (0.66-0.90)	0.85 (0.74-0.97)	0.002
Low C3 (<0.7 g/L)	133/623 (21.3)	26/78 (33.3)	107/545 (19.6)	0.006
C4 (g/L)	0.18 (0.15-0.22)	0.15 (0.11-0.19)	0.18 (0.15-0.22)	<0.001
Low C4 (<0.16 g/L)	216/623 (34.7)	43/78 (55.1)	173/545 (31.7)	<0.001
ESR (mm/h)	19.5 (10.3-36.8)	24.0 (13.0-45.5)	19.0 (10.0-34.0)	0.081
Elevated ESR (>20 mm/h)	293/600 (48.8)	41/73 (56.2)	252/527 (47.8)	0.181
ANA titres ≥ 1:320	186/621 (30.0)	16/78 (20.5)	170/543 (31.3)	0.052
RF positivity (>20 IU/mL)	276/603 (45.8)	34/75 (45.3)	242/528 (45.8)	0.935
Anti-SSA antibody positivity	409/621 (65.9)	58/78 (74.4)	351/543 (64.6)	0.091
Anti-Ro52 antibody positivity	340/621 (54.8)	41/78 (52.6)	299/543 (55.1)	0.678
Anti-SSB antibody positivity	171/621 (27.5)	13/78 (16.7)	158/543 (29.1)	0.022
Anti-CENPB antibody positivity	47/621 (7.6)	5/78 (6.4)	42/543 (7.7)	0.679
Anti-RNP antibody positivity	52/621 (8.4)	11/78 (14.1)	41/543 (7.6)	0.051
Anti-ACL antibodies positivity	25/471 (5.3)	7/70 (10.0)	18/401 (4.5)	0.108
Anti-β2GPI antibodies positivity	25/471 (5.3)	5/70 (7.1)	20/401 (5.0)	0.650

All values are presented as n (%) or median (interquartile). ITP: immune thrombocytopenia; C3: complement 3; C4: complement 4; ESR: erythrocyte sedimentation rate; ANA: antinuclear antibodies; RF: rheumatoid factor; anti-SSA: anti-Sjögren's syndrome A; anti-SSB: anti-Sjögren's syndrome B; anti-CENP-B: anti-centromere protein B; anti-RNP: anti-ribonucleoprotein; anti-ACL: anti-cardiolipin; anti-β2GPI: anti-β2-glycoprotein I.
*Mann-Whitney U-test for continuous data, Chi-squared test for compare binary data.

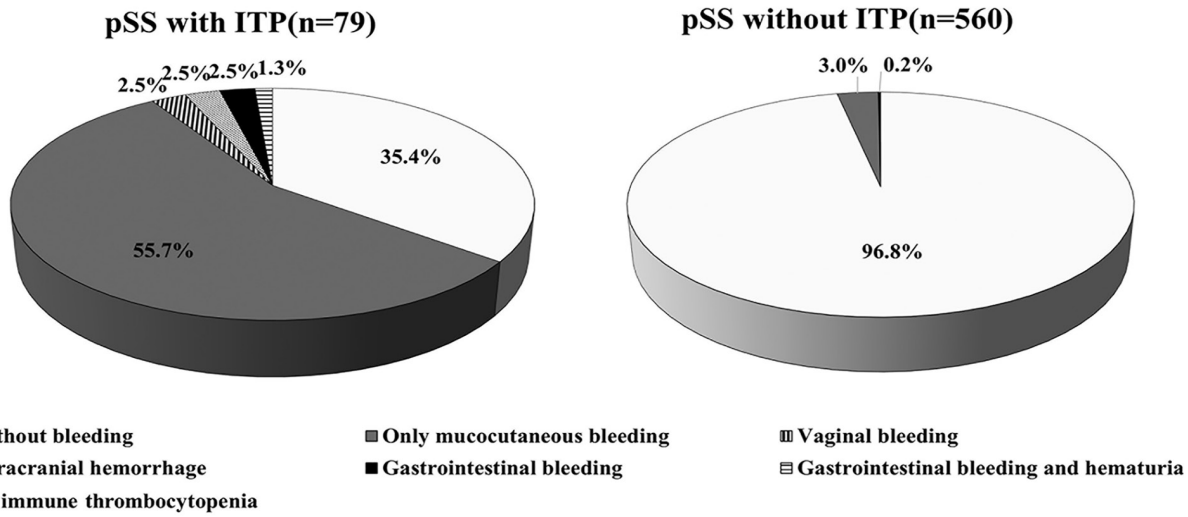

Fig. 2. Bleeding of pSS patients with secondary ITP.

to initially explored variables with possible statistical differences. Variables involving age, sex, disease duration, and those with a statistical *p*-value <0.1 in univariate analyses were included in multivariate regression analyses.

The results showed that lymphopenia was positively associated with secondary ITP in pSS patients (OR=3.154, 95% CI 1.185–8.395, *p*=0.021). Positive association was also detected between anaemia (OR=2.416, 95% CI 1.250–4.668, *p*=0.009), low C4 level (OR=2.904, 95% CI 1.563–5.394, *p*=0.001), or positive anti-RNP (OR=2.777, 95% CI 1.070–7.202, *p*=0.036) and ITP in patients with pSS. However, ILD (OR=0.429, 95% CI

Table III. Analysis of factors associated with secondary ITP among pSS patients.

Variables	OR	95%CI	*p*-value*
Age (years)	1.023	0.998-1.049	0.074
Gender (female)	2.610	0. 571-11.932	0.216
Disease duration (months)	0.999	0.996-1.003	0.657
Interstitial lung disease	0.429	0.203-0.907	0.027
Lymphopenia (<0.8×10^9/L)	3.154	1.185-8.395	0.021
Anaemia (haemoglobin <110 g/L)	2.416	1.250-4.668	0.009
Low C3 (< 0.7 g/L)	1.192	0.611-2.325	0.606
Low C4 (< 0.16 g/L)	2.904	1.563-5.394	0.001
ANA titres ≥ 1:320	0.469	0.221-0.996	0.049
Anti-SSA antibody positivity	1.884	0.900-3.778	0.095
Anti-SSB antibody positivity	0.288	0.126-0.658	0.003
Anti-RNP antibody positivity	2.777	1.070-7.202	0.036
Anti-ACL antibodies positivity	2.746	0.905-8.331	0.075

ITP: immune thrombocytopenia; C3: complement 3; C4: complement 4; ANA: antinuclear antibodies; anti-SSA: anti-Sjögren's syndrome A; anti-SSB: anti-Sjögren's syndrome B; anti-CENP-B: anti-centromere protein B; anti-RNP: anti-ribonucleoprotein; anti-ACL: anti-cardiolipin.
*Multivariate logistic regression analysis.

0.203-0.907, p=0.027) and ANA titres ≥1:320 (OR=0.469, 95% CI 0.221-0.996, p=0.049) were found negatively associated with secondary ITP in pSS patients, as well as the presence of positive anti-SSB (OR=0.288, 95% CI 0.126-0.658, p=0.003). Other factors such as sex, gender and disease duration were not significantly related to secondary ITP in patients with pSS.

Discussion

This is the first retrospective study with a large sample size to investigate clinical characteristics and relevant factors of secondary ITP in patients with pSS. The results showed that the prevalence of secondary ITP in patients with pSS was 12.4%, and 5.0% pSS patients had severe ITP, suggesting that secondary ITP was common in patients with pSS. The prevalence of ITP associated with pSS in our study is similar to a previous study (12.03%) involving Chinese patients (14). In contrast, we investigated the autoantibody profiles and relevant factors of secondary ITP in patients with pSS.

Our findings indicated that pSS patients with secondary ITP had higher proportion in bleeding than those without ITP; more than half of pSS patients with secondary ITP experienced mucocutaneous bleeding, but only 8.9% of them had visceral bleeding. These results are consistent with previous studies. A cross-sectional study enrolling 302 patients with ITP found slightly lower frequencies of mucocutaneous bleeding (51.3%) and severe bleeding (6.6%) in patients with ITP (18). In another small-scale study, 65.7% pSS patients with secondary ITP experienced bleeding symptoms, among which only 2.8% had severe haemorrhage (14). All these suggested that mucocutaneous bleeding is common but severe bleeding seems comparatively rare in pSS patients with secondary ITP. It seems reasonable to assume that patients who have severe bleeding may have lower levels of platelet than those with mild bleeding. However, we found that among pSS patients with secondary ITP, patients with severe ITP did not show higher proportion in severe bleeding than those with non-severe ITP. It has been reported that in ITP patients with lower platelet counts, increased platelet reactivity (platelet clotting capacity) was associated with decreased risk of bleeding (19). Thus, pSS patients with severe ITP who did not experience severe bleeding may have increased platelet reactivity. The relationship between platelet count and bleeding risk in pSS patients with secondary ITP need to be further explored.

We found some special clinical characteristics of pSS patients with secondary ITP compared with those without ITP. ILD is a main risk factor for poor prognosis of pSS (20), and the pathogenesis isn't fully understood (21). This study showed that ILD was negatively associated with secondary ITP in patients with pSS, indicating a possible role of platelets in the development of ILD. Additionally, the prevalence of ILD in our study (35.2%) is similar to a previous study (39.1%) from China (22), but higher than some previous reports (11%-20%) from European countries (23, 24), suggesting that the prevalence of ILD in patients with pSS is different in China and European countries.

Our data showed that pSS patients with secondary ITP had a higher proportion of lymphopenia, which was positively associated with the development of secondary ITP. Nevertheless, the underlying mechanisms need more studies to investigate. In addition, our data revealed that the haemoglobin level was lower in pSS patients with secondary ITP and anaemia was positively associated with the development of secondary ITP, suggesting certain potential co-influential factors such as serum iron. The role of iron in the formation and function of erythrocytes is well established. Iron levels direct the differentiation of megakaryocytic progenitors and contribute to energy production in platelet mitochondria (25). Therefore, iron may be a key link in thrombocytopenia and anaemia. More efforts should be devoted in exploring the underlying mechanisms and uncovering possible treatment targets in the future.

It has been reported that autoantibodies inducing complement-mediated destruction of platelets as well as inhibiting megakaryocyte function are involved in the development of ITP (26, 27). Nonetheless, the immunological characteristics of pSS patients with secondary ITP are still rarely documented. Our results suggested that positive anti-RNP and low level of C4 might be two potential risk factors for the development of secondary ITP, while ANA titres ≥1:320 and positive anti-SSB might be potential protective factors. A recent study also reported that complement levels are reduced in one-third of patients with ITP and low level of C4 could predict more severe ITP (28). Therefore, C4 seems play a key role in the development of ITP, indicating the potential of complement-directed therapies. This is an initial study exploring the role of anti-SSB

and anti-RNP in the development of ITP associated with pSS, more research is needed to verify our findings and explore the underlying mechanisms.

To date, the pathogenesis of pSS-associated thrombocytopenia has not yet been elucidated fully. Previous studies mainly focused on plasma P-selectin autoantibodies (29), recent studies pay more attention to Toll-like receptor and B cell activating factors (30). The new insights into pathogenesis of pSS-associated thrombocytopenia may inform potential treatment therapies. Currently, no specific therapeutic agent has been recommended to treat pSS-associated thrombocytopenia in clinical practice, although some evidence of potential drugs such as thrombopoietin receptor agonist and fostamatinib is available (31, 32). More high-quality studies are needed to explore potential safe and effective agents.

There are some limitations in this study. First, this is a single-centre retrospective study involving 639 Chinese patients with pSS, some potential confounders may be unknown although we adjusted main factors in our retrospective analyses. It provides a deeper understanding of factors associated with secondary ITP in Chinese patients with pSS, but the results may not be generalised to all ethnicities. Second, we were not able to analyse and report the outcomes of thrombocytopenia associated with pSS because we only retrospectively analysed the characteristics and factors at one time-point. In the future, multi-centre prospective studies with long term follow-up are needed to confirm our findings and explore the potential therapies for thrombocytopenia in patients with pSS.

Conclusions

Over 10% of patients with pSS had secondary ITP, among whom severe hemorrhage was uncommon. Lymphopenia and anaemia were positively related to secondary ITP, while ILD was negatively associated with secondary ITP. Positive anti-RNP and low level of C4 seem be two potential risk factors for secondary ITP in patients with pSS, but ANA titres ≥1:320 and positive anti-SSB may be potential protective factors. Future prospective studies are needed to verify the findings of this study.

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