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Anti-phospholipid antibody syndrome occurrence in patients with persistent anti-phospholipid antibodies

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Abstract

We investigated the overall frequency of anti-phospholipid syndrome (APS) occurrence in Korean patients with consecutively detected anti-phospholipid antibodies with an interval of 12 weeks (persistent aPLs). We retrospectively reviewed the results of blood tests of aPLs in 14,889 patients in whom aPL tests were performed at Yonsei University College of Medicine, Severance Hospital, from January 2012 to August 2018, and included 833 patients with persistent aPLs. We obtained clinical and laboratory data including anti-cardiolipin antibodies IgM and IgG, anti-beta2 glycoprotein1 IgM and IgG, and lupus anticoagulant (LAC). Of 833 patients with persistent aPLs, 96 patients (11.5%) had APS (84 patients had thrombotic events and 12 had pregnancy morbidity). Among aPLs, LAC was detected in patients with APS more frequently than asymptomatic carriers of aPLs (46.9% vs. 25.9%, $p < 0.001$). Patients with LAC (relative risk (RR) 2.558, $p < 0.001$) and aPLs ≥ 2 (RR 1.731, $p = 0.014$) exhibited the higher rate of APS occurrence than those without. Moreover, patients with aPLs ≥ 3 and aPLs ≥ 4 exhibited the higher rates of APS occurrence than those without (RR 2.753, $p < 0.001$ and RR 3.209, $p = 0.013$). Meanwhile, patients with ANA, anti-dsDNA, anti-SSA/Ro, and SLE exhibited the increased frequency of LAC positivity, compared to those without (RR 3.304, $p = 0.005$, RR 4.269, $p = 0.032$, RR 3.750, $p = 0.041$ and RR 8.828, $p < 0.001$, respectively). APS occurs in 11.5% of Korean patients with persistent aPLs. LAC positivity and aPLs ≥ 2 were significantly associated with APS occurrence. SLE and SLE-related autoantibodies were associated with LAC positivity.

Keywords Anti-phospholipid syndrome · Anti-phospholipid antibody · Risk factor · Asymptomatic carriers

Introduction

Anti-phospholipid syndrome (APS) is a systemic autoimmune disease, which is characterised by typical arterial and venous thrombosis or pregnancy morbidity by consecutively detected anti-phospholipid antibodies (aPLs) with an interval of 12 weeks or greater [1]. In this study, to clarify their clinical meaning, we named "consecutively detected aPLs with an interval of 12 weeks or greater" as "persistent aPLs". The classification of APS is based on the revised classification criteria for APS, which requires at least one clinical manifestation of thrombosis

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or pregnancy morbidity and one or more aPLs [2–4]. The revised classification criteria recognised 5 autoantibodies such as anti-cardiolipin antibodies (aCL) IgM and IgG, anti-beta2 glycoprotein 1 (aβ2GP1) IgM and IgG, and lupus anticoagulant (LAC) [2]. In the absence of clinical manifestations of APS, APS cannot be classified despite the presence of persistent aPLs. In real clinical settings, physicians occasionally encounter patients with persistent aPLs without clinical symptoms or signs of thrombosis. In cases of pregnancy morbidity such as recurrent spontaneous abortions, the classification of APS mainly depends on persistent aPLs, as patients are usually referred by obstetricians [5]. Whereas, in cases of arterial or venous thrombosis suspected, the classification of APS is often based on the confirmation of thrombosis by imaging, Doppler studies, or histopathology [2–4]. We defined patients, who have persistent aPLs, but exhibit no thrombotic events or pregnancy morbidity, as asymptomatic carriers of aPLs. So far, there have been several reports on the contributors to APS occurrence in patients with persistent aPLs such as LAC positivity, double or triple aPLs, and underlying autoimmune diseases [6, 7]. However, there was no report on predictors for APS occurrence in Korean patients with persistent aPLs. Hence, in this study, we investigated the overall frequency of APS occurrence and its predictors in Korean patients with persistent aPLs.

Materials and methods

Patients

We retrospectively reviewed the results of blood tests of aPLs in 14,889 patients, in whom tests of aPL were performed at Yonsei University College of Medicine, Severance Hospital, from January 2012 to August 2018. APS and other diseases such as comorbidities, autoimmune diseases, and concurrent malignancies were automatically extracted by Clinical Data Repository system of our institute and Diagnosis of diseases were identified in the 10th revised International Classification Diseases and medications administered were verified by the Korean Drug Utilisation Review system. Of 14,889 patients, 10,374 patients were excluded due to no aPLs detected at the first blood test. Of 4515 patients, 3672 patients were excluded due to aPLs negativity on the second blood test. The equipment to automatically measure the concentration aCL IgM/IgG and aβ2GP1 IgM/IgG was replaced with a new one since January 2012. Therefore, 10 patients, whose first results of aPLs were measured by the old equipment before January 2012 and who were diagnosed with APS after January 2012, were excluded to minimise the discordance between the two machines. Finally, we included 833 patients with persistent aPLs and reviewed their medical records. Of 833 patients, 96 patients were classified as APS based on the revised classification criteria for APS (Fig. 1) [2]. This study was approved by the Institutional Review Board (IRB) of Severance Hospital (4-2017-1254), and

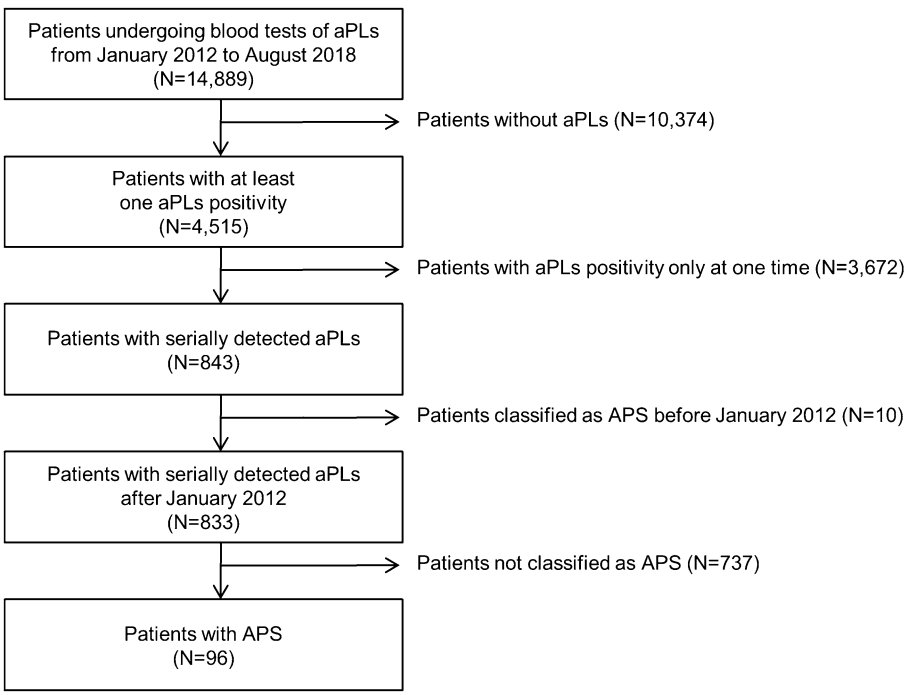


Fig. 1 Inclusion of patients with consecutively detected anti-phospholipid antibody. *APS* anti-phospholipid syndrome

the patient's written informed consent was waived by the approving IRB, as this was a retrospective study.

Measurement of aPLs

Anti-phospholipid antibodies including aCL IgM/IgG and aβ2GP1 IgM/IgG were tested by the automated immunoassay equipment, Phadia 250 (Phadia, Sweden), using fluorescence enzyme immunoassay (FEIA) method. The positivity of both aCL and aβ2GP1 was based on testing for aCL IgG and IgM, aβ2GP1 IgG and IgM and LA on two or more consecutive occasions at least 12 weeks apart. In cases of patients suspected of catastrophic APS with aPLs positivity on two timepoints with an interval of fewer than 12 weeks, treatments were immediately initiated and tests of APS were repeated 12 weeks after the first sPLs' positivity. According to Sapporo classification criteria, laboratory criteria were described as the following: (i) LAC positivity on ≥ 2 occasions at least 12 weeks apart; (ii) aCL (IgG and/or IgM) in medium or high titre (i.e., > 40 or above the 99th percentile), on two or more occasions at least 12 weeks apart; and (iii) aβ2GP1 (IgG and/or IgM) in medium or high titre (i.e., above the 99th percentile) on two or more occasions at least 12 weeks apart. 99th percentile of the results may be different according to study population. Therefore, the presence of aCL and aβ2GP1 was set when its titre was in moderate and high (> the 99th percentile) according to the recommendation of the manufacturer after the validation with Korean people [2, 8]. LAC was screened and confirmed by dilute Russell viper venom time (Instrumentation Laboratory, MA, USA) [9]. Since the results aCL IgM/IgG and aβ2GP1 IgM/IgG did not come from a normal distribution in the Shapiro–Wilk test, we used categorical data (presence or absence) rather than continuous data. We assume that the large numbers of patients without aPLs may contribute to no normal distribution.

Clinical data and autoantibodies

We obtained age at diagnosis, gender, and gap time and follow-up duration as demographic data. The follow-up duration was defined as a period from the second detection of aPLs to the last visit to our institute. Gap time was defined as a period from the second detection of aPLs with an interval of 12 weeks or greater to the classification of APS. In 96 patients with APS, we reviewed clinical manifestations related to APS such as deep vein thrombosis (DVT), pulmonary thromboembolism (PTE), myocardial infarction (MI) or angina, stroke, transient ischaemic attack (TIA), peripheral arterial thrombosis (PAT), and pregnancy morbidity [2, 3, 10]. We also reviewed anticoagulants and anti-platelet agents administered after diagnosis of APS such as warfarin, rivaroxaban, aspirin, clopidogrel, and cilostazol [11]. In addition, we reviewed the medical records regarding comorbidities which are known as perpetrators of atherosclerosis, such as hypertension, diabetes mellitus, and dyslipidaemia [9, 12]. In addition, we searched concurrent autoimmune connective tissue diseases including systemic lupus erythematosus (SLE), Sjogren syndrome (SS), and systemic sclerosis (SSc) and concurrent malignancies. Autoantibodies along with APLs were also investigated [13, 14]. Because not all patients underwent blood tests for autoantibodies, we unified the calculation strategy that denominator is the number of patients undergoing a test for each autoantibody and numerator is the number of patients having a positive result.

Statistical analyses

All statistical analyses were conducted using SPSS software (version 23 for windows; IBM Corp., Armonk, NY, USA). Continuous variables were expressed as the median [interquartile range, IQR] and categorical variables were expressed as number and the percentage. Significant differences in the categorical variables between the two groups were analysed using the Chi-square and Fisher's exact tests, and significant differences in the continuous variables between the two groups were compared using the independent samples *t* test. The relative risk (RR) of each variable for either APS occurrence or LAC positivity was analysed by contingency tables and the Chi-square test. We also obtained the sensitivity, the specificity, the positive predictive value (PPV), and the negative predictive value (NPV) by contingency tables. *p* values less than 0.05 were considered statistically significant.

Results

Comparison of baseline variables between patients with and without APS

We divided 833 patients having persistent aPLs into two groups based on the classification of APS, and compared demographic data, comorbidities and APLs between the two groups. Of 833 patients with persistent aPLs, the overall frequency of APS occurrence was 11.5%. There were no differences in age at diagnosis, gender, follow-up duration, and comorbidities such as hypertension, diabetes mellitus, and dyslipidaemia between the two groups. Among aPLs, LAC was detected in patients with APS more frequently than asymptomatic carriers of aPLs [45 of 96 APS patients (46.9%) vs. 189 of 737 asymptomatic carriers (25.6%), $p < 0.001$]. aCL IgM, aCL IgG, aβ2GP1 IgM, and aβ2GP1 IgM were not significantly different between carriers and APS patients. In addition, patients with APS exhibited the higher numbers of aPLs positivity than asymptomatic

carriers of aPLs (38.5% vs. 26.6% for 2 or greater, $p=0.014$, 18.8% vs. 7.7% for 3 or greater, $p<0.001$ and 6.3% vs. 2.0% for 4 or greater, $p=0.013$) (Table 1).

Relative risk of variables for APS occurrence

We obtained RR for APS occurrence using variables with statistical significance in the comparison analysis between asymptomatic carriers of aPLs and patients with APS. Patients with LAC exhibited the higher rate of APS occurrence than those without (RR 2.558, 95% confidence interval (CI) 1.658, 3.947, sensitivity 46.9%, specificity 74.4%, PPV 19.2%, and NPV 91.5%). Furthermore, patients with more than 2 aPLs showed the higher frequency of APS occurrence that those without (RR 1.731, 95% CI 1.112, 2.694, sensitivity 38.5%, specificity 73.4%, PPV 1.9%, and NPV 90.2%). In addition, regardless of types of aPLs, patients with more than 3 aPLs and 4 aPLs showed the increased rates of APS occurrence, compared to those without (RR 2.753, 95% CI 1.542, 4.914 and RR 3.209, 95% CI 1.214, 8.480, respectively) (Fig. 2).

Relative risk for APS occurrence based on the number and type of aPLs

To investigate an RR of each number of aPLs positivity compared to 1+ aPLs, we divided all subjects into five groups based on the number of positive aPLs such as 1+ to 5+. We obtained relative risks of 2+ to 5+ for APS in comparison with 1+. When the numbers of sPLs were 2+, 3+, 4+, and 5+, relative risks were 1.253 ($p=0.420$), 2.620 ($p=0.005$), 2.620 ($p=0.219$), and 4.585 ($p=0.008$), respectively. 2+ exhibited no increased risk for APS compared to 1+. Whereas, 4+ exhibited an apparent difference compared to 1+, but there was no statistical significance due to a small number of subjects with four positive aPLs. Although there was no significance in the comparison analysis between 1+ and 4+, as the number of positive aPLs increased, the proportion of APS also increased (Fig. 3). This result is in the line with the result of Fig. 2. Thus, we assume that in real clinical practice, it might be reasonable to use relative risks of ≥ 2, ≥ 3, or ≥ 4 aPLs, similar to each RR of 2+ to 5+ aPL compared to 1+ aPL.

Characteristics of 96 patients with APS

The mean age was 40.5 years and 53 patients (55.2%) were women. The mean gap time and follow-up duration were 3.8 and 31.3 months. The most frequently consecutively detected aPL was LAC (46.9%) among aPLs and stroke was the most common thrombotic event (31.3%). Twelve women (12.5%) were classified as APS due to pregnancy morbidity [2, 3]. Anticoagulants and anti-platelet agents, hydroxychloroquine, concurrent diseases, and autoantibodies are described in Table 2.

Table 1 Comparison of baseline variables between patients with and without APS

Variables	Asymptomatic carriers of aPLs ($N=737$)	Patients with APS ($N=96$)	p value
Demographic data			
Age at diagnosis (years)	39.0 (14.0)	37.5 (21.0)	0.663
Female gender [N, (%)]	410 (55.6)	53 (55.2)	0.938
Follow-up duration (months)	28.0 (38.0)	25.0 (39.0)	0.577
Comorbidities [N, (%)]			
Hypertension	180 (24.4)	21 (21.9)	0.583
Diabetes mellitus	71 (9.6)	8 (8.3)	0.683
Dyslipidaemia	165 (22.4)	22 (22.9)	0.907
Presence of aPLs			
aCL IgM	282 (38.3)	32 (33.3)	0.348
aCL IgG	281 (38.1)	42 (43.8)	0.288
aβ2GP1 IgM	110 (14.9)	14 (14.6)	0.929
aβ2GP1 IgG	151 (20.5)	28 (29.2)	0.052
LAC	189 (25.6)	45 (46.9)	< 0.001
Number of aPLs ≥ 2	196 (26.6)	37 (38.5)	0.014
Number of aPLs ≥ 3	57 (7.7)	18 (18.8)	< 0.001
Number of aPLs ≥ 4	15 (2.0)	6 (6.3)	0.013

Values are expressed as median [interquartile range, IQR] or N (%)

APS anti-phospholipid syndrome, *aPL* anti-phospholipid antibody, *aCL* anti-cardiolipin, *aβ2GP1* anti-beta2 glycoprotein 1, *LAC* lupus anticoagulant

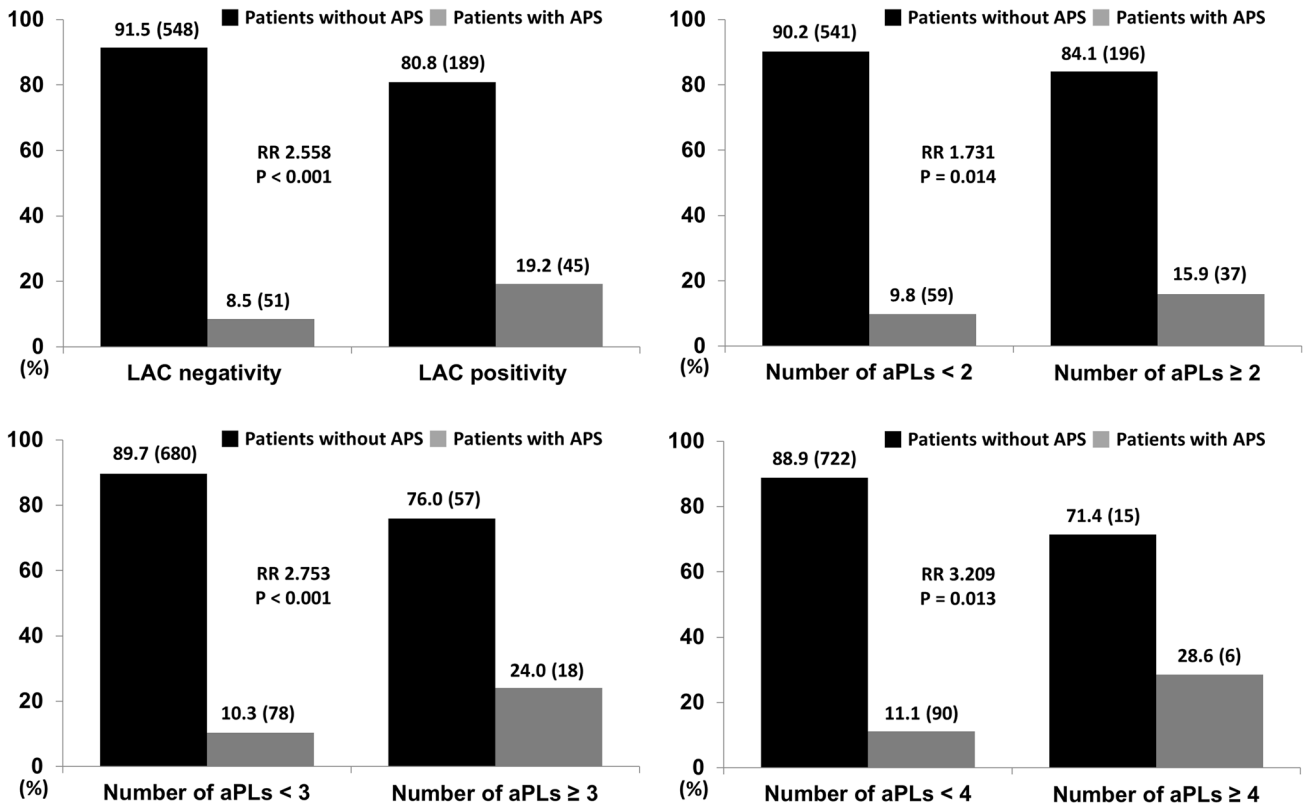


Fig. 2 Association of aPLs and APS occurrence. Values are expressed as a percentage and (the number of patients). LAC positivity and the number of aPL more than 2, 3, and 4 were significantly associated with APS occurrence. *aPL* anti-phospholipid antibody, *APS* anti-phospholipid syndrome, *LAC* lupus anticoagulant

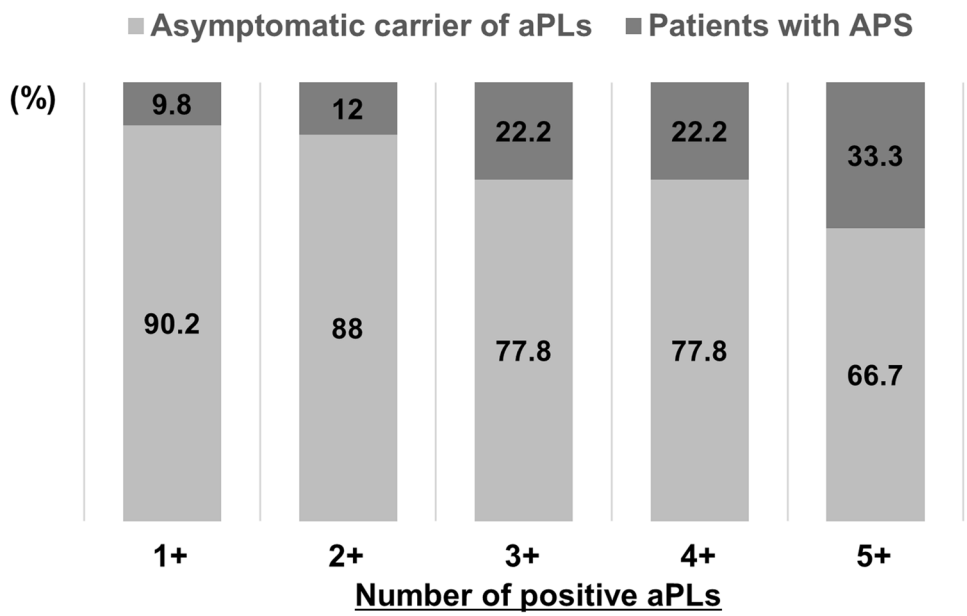
Relative risk of autoantibodies and autoimmune connective tissue diseases for LAC positivity

Because LAC appear to be a predictor for APS occurrence among aPLs (Table 1 and Fig. 2), we analysed the association of autoantibodies with LAC positivity. Patients with antinuclear antibody (ANA) exhibited the higher rate of LAC positivity than those without (RR 3.304, 95% CI 1.411, 7.736). Furthermore, patients with anti-ds DNA and those with anti-SSA/Ro exhibited the increased frequency of LAC positivity, compared to those without (RR 4.269, 95% CI 1.053, 17.306 for anti-ds DNA and RR 3.750, 95% CI 1.007, 13.965 for anti-SSA/Ro). SLE also significantly contributed to LAC positivity (RR 8.828, 95% CI 2.367, 32.929) (Fig. 4).

Discussion

In this study, the overall frequency of APS occurrence was 11.5% in Korean patients with persistent aPLs. Therefore, a majority of patients with persistent aPLs might not be classified as APS. However, we wonder whether these asymptomatic carriers of aPLs may be free from APS occurrence during follow-up. A previous study using data from the Register for Coagulation Disorders of the Finnish Red Cross Blood Service reported that 110 of 119 patients, who had been classified as asymptomatic carriers of aPLs, did not experience thrombotic events. Only 7.5% of asymptomatic carriers of aPLs were reclassified as APS at a mean follow-up duration of 7.2 years after aPLs detection [10]. Another prospective study reported that no episode of thrombosis was confirmed in any asymptomatic carriers of aPLs during a total of 36 months, although prophylactic anticoagulation was transiently administered for the period when subjects were at increased risk of thrombosis [15]. In this study, the mean follow-up time of asymptomatic carriers of aPLs was 32.6 months, which was shorter that of a previous study (2.7 years vs. 7.2 years). Therefore, if we follow up these subjects for 4.5 years or greater, we could approximately compare the incidence of thrombotic events in asymptomatic carriers of aPLs with that of the previous study [7].

In the present study, LAC positivity and the number of aPLs positivity ≥ 2 were significantly associated with APS occurrence in all Korean subjects with persistent aPLs. Although several previous studies insisted that LAC



Relative risk for APS (1+ aPLs as a reference)			
Number of positive aPLs	Relative Risk	95% confidence interval	P value
2+	1.253	0.723, 2.171	0.420
3+	2.620	1.370, 5.252	0.005
4+*	2.620	0.532, 12.902	0.219
5+	4.585	1.340, 15.685	0.008

Fig. 3 Relative risk for APS occurrence based on the number and type of aPLs. Values are expressed as a percentage. Asterisk indicates the number of subjects with 4 positive aPLs was too small to obtain a statistical significance despite a difference between the two groups. *aPL* anti-phospholipid antibody, *APS* anti-phospholipid syndrome, *aCL*: anti-cardiolipin antibodies, *aβ2GP1* anti-beta2 glycoprotein 1

positivity and double or triple aPLs should be closely associated with APS occurrence [6, 7], their contributions to APS occurrence still remain controversial. A previous study reported that the types of aPLs or the number of aPLs positivity did not differ between asymptomatic carriers with and without thrombotic events during follow-up [10]. Meanwhile, another study provided a result that LAC was detected in patients with both primary and secondary APS significantly more frequently than asymptomatic carriers of aPLs (46.9% vs. 37.6%, $p < 0.05$) [15]. In addition, there was a difference in the rate of LAC positivity between Western countries and Korea (56.0% in Finland or 46.9% in Spain vs. 28.1% in Korea) [10, 15]. In addition, in 96 patients with APS, LAC was the most common APL. Thus, although there were differences in detection assay methods between studies, we suggest that the clinical significance of persistent aPLs and the types of aPLs in asymptomatic carriers should be reconsidered in different ethnic and geographical backgrounds.

In 96 patients with APS, we assessed the association of concurrent diseases with APS occurrence or LAC positivity in three ways. First, in terms of risk factors for atherosclerosis, the frequency of hypertension, diabetes, and dyslipidaemia did not differ between patients with APS and asymptomatic carriers of aPLs, similar to a previous study [15]. We assume that no contribution of these risk factors might be due to a younger age of the study subjects than the general population vulnerable to atherosclerosis. Second, in terms of autoimmune connective tissues, SLE was associated with LAC positivity. Since LAC positivity was only a predictor for APS occurrence among aPLs, SLE could be considered associated with APS occurrence, which was in line with a previous study [10]. This association might depend on medical situations that APS may occur in 20–30% of SLE patients [16], and tests of aPLs may be performed in patients suspected of SLE more commonly than those not. Third, in terms of autoantibodies, LAC positivity was associated with ANA, anti-ds DNA and anti-SSA/Ro, which are frequently found in SLE patients. A majority of patients with persistent aPLs turned out to be asymptomatic carriers of aPLs, but the overall frequency of APS occurrence (11.5%) is not ignorable. Therefore, the regular and dynamic follow-up of

Table 2 Characteristics of 96 patients with APS

Variables	Values
Demographic data	
Age at diagnosis (years)	37.5 (21.0)
Female gender [*N*, (%)]	53 (55.2)
Gap time* (months)	0 (2.0)
Follow-up duration** (months)	25.0 (39.0)
aPLs [*N*, (%)]	
aCL IgM	32 (33.3)
aCL IgG	42 (43.8)
aβ2GP1 IgM	14 (14.6)
aβ2GP1 IgG	28 (29.2)
LAC	45 (46.9)
Clinical manifestations of APS [*N*, (%)]	
Stroke	30 (31.3)
DVT	24 (25.0)
PAT	14 (14.6)
PTE	13 (13.5)
Pregnancy morbidity	12 (12.5)
MI or angina	9 (9.4)
TIA	1 (1.0)
Number of patients with 2 thrombotic sites	7 (7.3)
Anticoagulants and anti-platelet agents [*N*, (%)]	
Warfarin	38 (39.6)
Aspirin	30 (31.3)
Rivaroxaban	12 (12.5)
Clopidogrel	11 (11.5)
Cilostazol	3 (3.1)
None	5 (5.2)
Hydroxychloroquine [*N*, (%)]	36 (37.5)
Concurrent diseases [*N*, (%%)]	
Autoimmune connective tissue diseases	
SLE	19 (19.8)
SS	1 (1.0)
SSc	1 (1.0)
Malignancies	
Breast cancer	2 (2.1)
Meningioma	1 (1.0)
Autoantibodies [*N*, (%)]	
Antinuclear antibody (*N*=96)	39 (40.6)
Anti-ds DNA (*N*=75)	12 (16.0)
Anti-RNP (*N*=53)	5 (9.4)
Anti-Sm (*N*=57)	5 (8.8)
Anti-SSA/Ro (*N*=61)	13 (21.3)
Anti-SSB/La (*N*=60)	3 (5.0)
Anti-Scl70 (*N*=41)	1 (2.4)
Anti-citrullinated peptide antibody (*N*=41)	0 (0)
Anti-centromere (*N*=30)	1 (3.3)
MPO-ANCA (or P-ANCA) (*N*=72)	2 (2.8)
PR3-ANCA (or C-ANCA) (*N*=71)	0 (0)

Values are expressed as median [interquartile range, IQR] or *N* (%)

APS anti-phospholipid syndrome, *aPL* anti-phospholipid antibody, *aCL* anti-cardiolipin, *aβ2GP1* anti-beta2 glycoprotein 1, *LAC* lupus anticoagulant, *DVT* deep vein thrombosis, *PTE* pulmonary thromboembolism, *MI* myocardial infarction, *TIA* transient ischaemic attack, *PAT* peripheral arterial thrombosis, *SLE* systemic lupus erythematosus, *SS* Sjogren syndrome, *SSc* systemic sclerosis, *MPO* myeloperoxidase, *ANCA* antineutrophil cytoplasmic antibody, *P* perinuclear, *PR3* proteinase 3, *C* cytoplasmic

*Gap time: a period from the second detection of aPLs to diagnosis of APS

**Follow-up duration: a period from the second detection of aPLs to the last visit

asymptomatic carriers of aPLs, particularly those who have LAC or are suspected of SLE or classified as SLE, should be done.

In this study, we first investigated the overall frequency of APS occurrence and its predictors in Korean subjects with persistent aPLs. However, this conclusion was possible, because this study was a retrospective study. Since our institute is one of the biggest tertiary hospitals in Korea, a considerable number of patients are being referred to our institute from outside hospital. For this reason, the severity of patients with thrombotic events or pregnancy morbidity is considered very high. Therefore, a test for APS has become a part of the initial essential tests in our institute. These situations might increase the proportion of asymptomatic carriers of aPLs and they might also leave an ethical issue of the indiscriminate evaluation of sPLs. Therefore, we could not clarify how many patients of 14,889 patients actually had suspicion of APS or why tests for APS were performed.

Our study also has several issues. Due to a retrospective study design of this study, we could not gain direct information on atherosclerosis, which is one of the risk factors for thrombotic events. Furthermore, because the reasons to perform tests for aPLs were documented in a fourth of asymptomatic carriers of aPLs approximately, we could not compare the reasons for test performance between APL patients and asymptomatic carriers. The follow-up duration was too short to examine the natural course of asymptomatic carriers of aPLs. In addition, the total number of this monocentric study is not small; however, it is not as large as a nationwide study to represent the clinical features of both patients with APS and asymptomatic carriers of aPLs in Korea. If we prospectively enrol more patients with persistent aPLs from nationwide multi-centres and follow-up asymptomatic carriers of aPLs longer, we believe that we could clearer demonstrate the major predictors for APS occurrence and their related factors in Korean patients with persistent aPLs. In conclusion, the overall frequency of APS occurrence was 11.5% in Korean patients with persistent aPLs. LAC positivity and the number of aPLs more than 2 were significantly associated with APS occurrence. SLE and SLE-related autoantibodies were associated with LAC positivity.

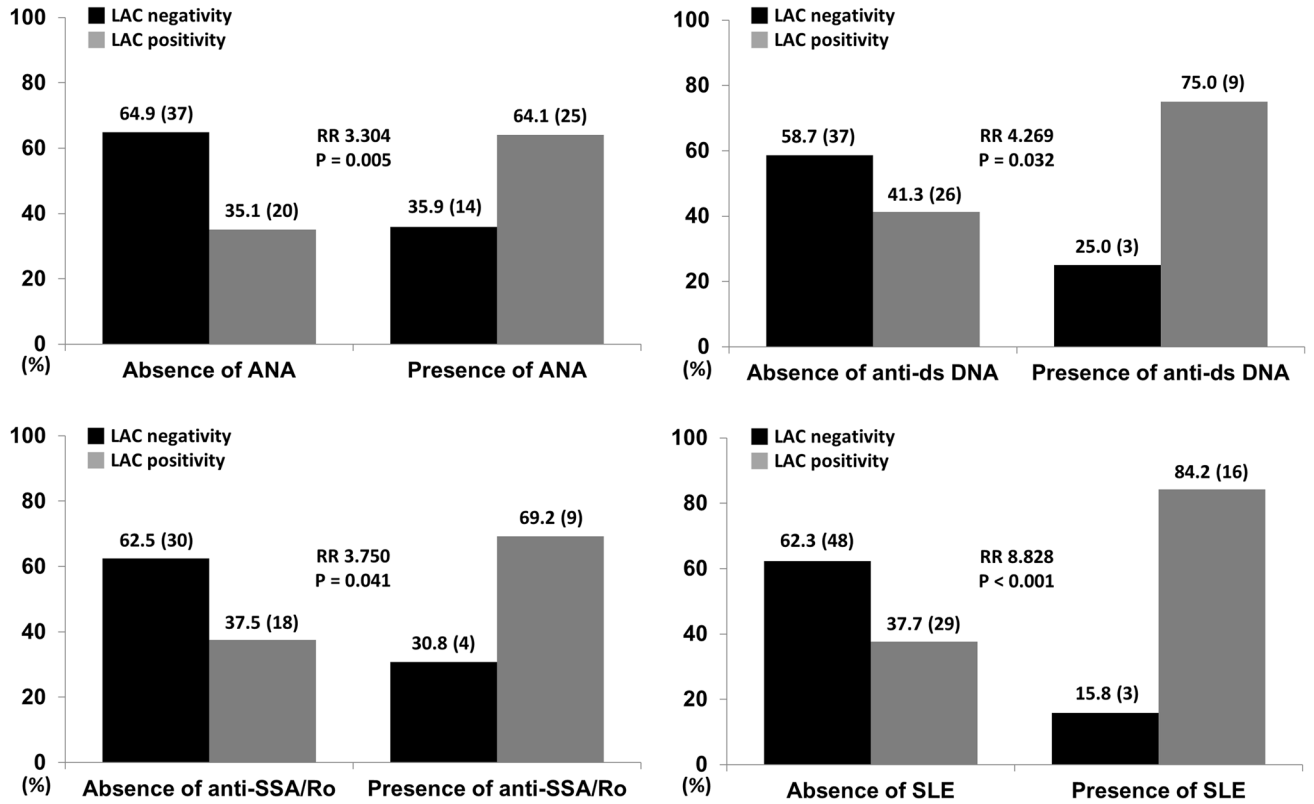


Fig. 4 Association of autoantibodies and autoimmune disease with LAC positivity. Values are expressed as a percentage and (the number of patients). ANA, anti-ds DNA, and anti-SSA/Ro were significantly associated with LAC positivity. SLE also contributed to LAC positivity. *LAC* lupus coagulant, *SLE* systemic lupus erythematosus

Author contributions HC and SWL collected the data and analysed the results and wrote the manuscript, under the guidance of SSA and JJS, and YBP contributed to writing and critically reviewed the manuscript. HC and SWL designed, analysed the results, critically reviewed the manuscript, and wrote the final version. All authors read and approved the final manuscript.

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Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Ethical approval All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards. This study was approved by the Institutional Review Board (IRB) of Severance Hospital (4-2017-1254).

Informed consent The patient's written informed consent was waived by the approving IRB, as this was a retrospective study.

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