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ORIGINAL ARTICLE

Non-criteria autoantibodies in antiphospholipid syndrome may be associated with underlying disease activity

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Abstract

Background Antiphospholipid syndrome (APS) is a systemic autoimmune disorder characterized by persistent antiphospholipid antibodies (aPLs) with arterial and venous thrombosis and/or pregnancy morbidity. In recent years, several studies have highlighted the potential role of non-criteria aPL in diagnosing APS patients.

Aim This study aimed to determine the association of the presence of non-criteria aPL antibodies to the clinical and laboratory features of patients with a diagnosis of APS.

Methods Eighty patients diagnosed with APS and under observation in the rheumatology clinic of Ankara City Hospital were assessed. Patient demographic and clinical features were meticulously recorded. Non-criteria antibodies tested in our center included antiphosphatidylserine IgA, antiphosphatidylserine IgM, beta 2 glycoprotein IgA, anti-cardiolipin IgA, antiphospholipid antibody IgG, and antiphospholipid antibody IgM. Antibodies from patients who were tested for at least one non-criteria antibody were documented.

Results Out of 80 patients, 55 (68.8%) were tested for at least one non-criteria antibody, and 29 of those patients (52.7%) tested positive for at least one non-criteria antibody. The antiphospholipid antibody IgM and the beta 2 glycoprotein IgA were the most commonly tested non-criteria antibodies. Patients with non-criteria antibody positivity had a higher frequency of Ds DNA positivity and low complement (62.0% vs. 35.0%, $p=0.042$; 69.0% vs. 38.0%, $p=0.023$), respectively. In addition, positivity for anti-cardiolipin IgG and b2 glycoprotein IgG was significantly higher in the group positive for non-criteria antibodies (79% vs. 31%, $p\leq0.001$; 72.0% vs. 19%, $p\leq0.001$). There was no significant difference between the clinical features of patients with at least one positivity for non-criteria antibodies and those without.

Conclusion Systemic lupus erythematosus (SLE) is the most commonly associated disease with APS, being present in approximately 35% of cases [1]. Since the majority of the patient group in our study had APS that was secondary to SLE, non-criteria antibody positivity may be linked to the immunological activity of SLE. Large multicenter studies are necessary to investigate the clinical significance of isolated/combined positivity for criterion/non-criteria aPLs.

Keywords Antiphospholipid syndrome · Auto-immune disorder · Non-criteria antibody

Introduction

Antiphospholipid syndrome (APS) is an autoimmune disease that is characterized by the presence of circulating antiphospholipid antibodies (aPL). This condition is known to cause a range of vascular and obstetric manifestations due to the activation of thrombotic and inflammatory mechanisms [2]. The clinical manifestations of antiphospholipid syndrome (APS) commonly encompass venous thromboembolism, stroke, recurrent early miscarriages, and late pregnancy losses, as reported in the literature [3].

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As per the 2006 APS classification criteria, also known as the Sydney criteria, the diagnosis of APS requires the presence of at least one clinical and one laboratory criteria. Laboratory criteria consist of the existence of lupus anticoagulant (LA), high levels of anti-cardiolipin (aCL), and anti-β2 glycoprotein-I (aβ2GPI) immunoglobulin isotype G (IgG) or M (IgM) [3]. The detection of antibodies, a crucial component of the diagnostic process, necessitates their identification on at least two separate occasions, with a minimum interval of 12 weeks.

According to the Sydney classification criteria, the clinical arm refers to thrombosis and pregnancy morbidity, which refers to the occurrence of at least three consecutive miscarriages before the 10th week of gestation; one or more fetal losses after the 10th week of gestation; stillbirth, premature, and severe preeclampsia (PE); or prematurity caused by placental insufficiency [3].

Hughes and Khamashta (2003) introduced the concept of seronegative antiphospholipid syndrome (SN-APS). The SN-APS pertains to persons who exhibit clinical symptoms that strongly suggest a diagnosis of APS yet consistently yield negative results for aPL criteria [4].

In addition to the customary standards, various clinical and laboratory features were found to be associated with APS in numerous studies, such as heart valve disease, thrombocytopenia, neurological symptoms, aCL or aβ2GpI IgA, and anti-phosphatidylserine-prothrombin (aPS/PT) [5].

In recent years, numerous studies have emphasized the potential role of non-criteria aPLs in diagnosing APS patients [6]. Studies have demonstrated the presence of more than 30 non-criteria aPL in APS [7–9]. The aPS/PT and aβ2GPI domain I are considered "first-line" non-criteria aPLs [10], and the non-criteria antibody, which is included in GAPSS and APL-S for risk stratification in APS patients, is also associated with APS [11, 12].

In addition to diagnosing APS, the evaluation of non-criteria aPLs can contribute to the risk assessment for prognosis and associated clinical manifestations [13]. Utilizing non-criteria aPLs may prove beneficial in confirming or ruling out an elevated risk profile in individuals with deficient antibody profiles [14].

Notable evidence has been reported regarding the association between anti-phosphatidylserine/prothrombin (aPS/PT) antibodies and thrombosis and pregnancy morbidities [15–17]. Furthermore, aAnxV and aPS/PT have consistently received attention in recent years [5]. Antibodies against beta2-glycoprotein I (aDI) [18] and vimentin/cardiolipin (Vim/CL) have been identified as potential targets for APS [19]. APhL has been associated with arterial thrombosis and pregnancy-related morbidity [20].

The place and clinical relevance of non-criteria aPLs remain controversial. Most existing studies have evaluated only one or a few non-criteria aPLs, utilizing different diagnostic tests and study designs. Studies have focused on the clinical significance of these antibodies in seronegative APS patients. We aimed to determine the association of non-criteria aPL antibodies to APS patients' clinical and laboratory features.

Materials and method

We enrolled patients diagnosed with primary or secondary antiphospholipid syndrome according to the 2006 Sydney APS criteria [3] and undergoing regular monitoring at the rheumatology clinic of Ankara City Hospital. We retrospectively reviewed patients' charts to document the demographic characteristics, clinical manifestations, laboratory results, and imaging findings. We also record the various aspects of patients' medical conditions, including the duration of their illnesses, symptoms, coexisting ailments, and thrombosis locations as well as pregnancy losses, preeclampsia, and preterm labor of obstetric APS patients. We also noted whether patients had hemolytic anemia, leukopenia, thrombocytopenia, positive anti-nuclear antibody (ANA), positive double-stranded DNA (dsDNA), low complement levels, and a positive Coombs test. Every patient underwent the evaluation of LA, aCL IgG, aCL IgM, aβ2GPI IgG, and aβ2GPI IgM. Also, antiphosphatidylserine IgA, antiphosphatidylserine IgM, antiphosphatidylserine IgG, aβ2GPI IgA, aCL IgA, antiphospholipid antibody IgG, and antiphospholipid antibody IgM were the other antibodies that can be tested. The antibodies of the patients who underwent testing for at least one non-criteria antibody were recorded, as shown in Fig. 1. The patients diagnosed with APS and underwent non-criteria antibody testing were categorized into two groups: those with at least one positive non-criteria antibody and those with negative. The clinical and laboratory features of both groups were recorded.

Statistical Package for the Social Sciences (SPSS) version 22 was used to run the statistical analysis (IBM Corp., Armonk, NY). Shapiro–Wilk analysis was performed to determine the normality of the variables in addition to plots and histograms. Continuous variables were presented as median (interquartile range [IQR]) or as mean ± standard deviation according to normality. Comparisons of continuous variables between different groups were made by Mann–Whitney-U or Student t tests in accordance with normality. Categorical variables were expressed as numbers and percentages and compared by $\times 2$ test. In all analyses, p values < 0.05 were considered statistically significant.

Ethics approval (E1-22–2828) was obtained from the ethics committee of Ankara City Hospital. The study was conducted in accordance with the ethical standards of the 1964 Declaration of Helsinki and its subsequent amendments.

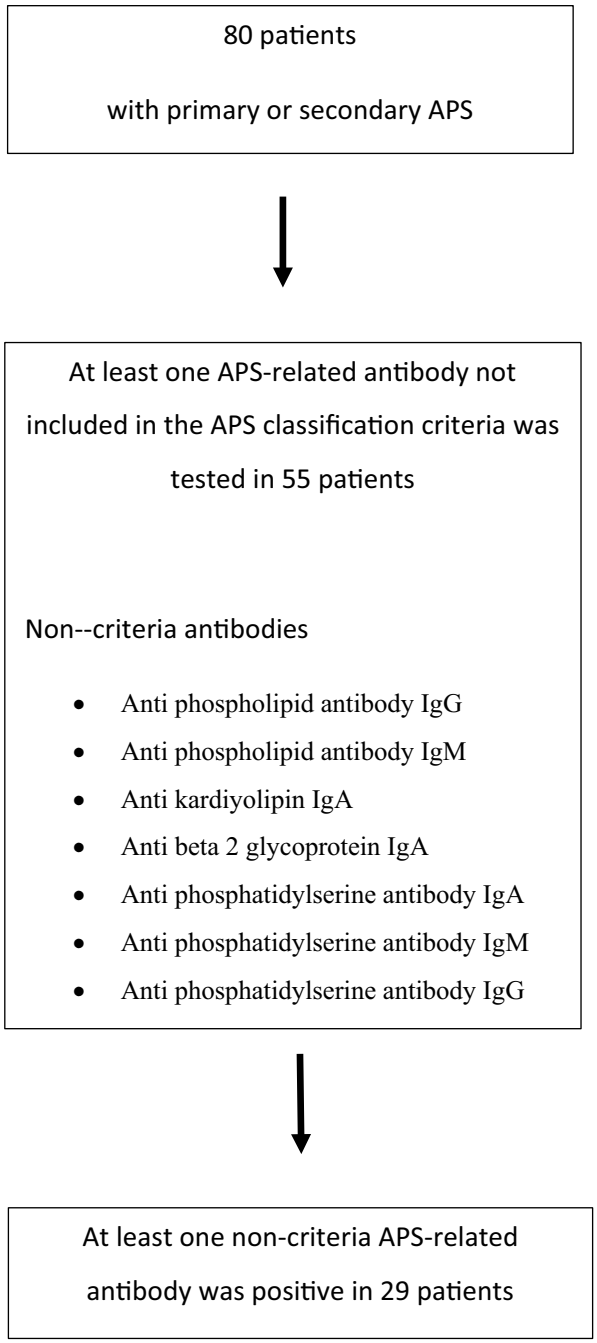

Fig. 1 Flow chart showing the patient evaluation process for recruitment

Results

The study comprised a total of 80 patients who were monitored for APS at our rheumatology clinic. Among these patients, 23 were diagnosed with obstetric APS, 39 with thrombotic APS, and 18 with thrombotic + obstetric APS. The predominant comorbidity noted in secondary APS patients was systemic lupus erythematosus (SLE), constituting 57 (71.2%) of cases. Table 1 displays the patients' demographic, clinical, and laboratory characteristics and compares the laboratory and clinical features of patients who underwent at least one non-criteria antibody test and those who did not undergo such testing. No discernible difference was observed in the clinical and laboratory characteristics of the aforementioned groups. In 68.8% of the patients, there was a minimum of one non-criteria antibody examined. Among these patients, 52.7% had at least one positive non-criteria antibody. The antiphospholipid antibody IgM ($n=40$) and beta 2 glycoprotein IgA ($n=35$) were the most frequently tested ones. The most common antibodies meeting the established criteria were LAC ($n=55$) and aCL IgG ($n=43$). According to the study results, positivity for non-criteria antibodies was significantly linked to higher dsDNA positivity and lower complement levels (62.0% vs. 35.0% $p=0.042$; 69.0% vs. 38.0%, $p=0.023$). Additionally, the occurrence of aCL IgG and aβ2GPI IgG positivity was significantly more common in the group positive for non-criteria antibodies when compared to those negative for non-criteria antibodies, respectively (79% vs. 31%, $p\leq0.001$; 72.0% vs. 19%, $p\leq0.001$). The study compared patients who have at least one non-criteria antibody positivity with those who have not in terms of other clinical and laboratory characteristics. The results are presented in Table 2, which shows no significant statistical difference between the two groups.

Discussion

The range of medical conditions addressed at the APS clinic is extensive. The investigation of the contribution of non-criteria antibodies, which have recently garnered significance in disease assessment, alongside the diagnostic antibodies, has been a subject of scholarly attention. The present study aimed to assess the association between non-criteria antibodies and clinical and laboratory features in individuals diagnosed with APS. Additionally, the study aimed to investigate the effect of patients' clinical and laboratory characteristics on the requisition of non-criteria antibodies. The present investigation documented an elevated frequency of dsDNA positivity and reduced complement levels among individuals exhibiting non-criteria aPL antibody positivity. The results of our study indicate that patients with secondary APS may exhibit antibody positivity that is not related to the diagnostic criteria, particularly in relation to the immunological mechanisms of the underlying primary disease.

Recognizing clinical indicators or ascertaining patients who lack antibodies remains a crucial task owing to the elevated rates of mortality and morbidity observed in APS patients. The diagnosis of a significant proportion (60.9%)

Table 1 Demographics, clinical and laboratory characteristics in patients

	N = 80	Non_criteria antibody tested (n:55)	Non criteria antibody untested (n:25)	p
Age, years, mean ± SD	42.7 ± 11.5	42.7 ± 11.5	42.8 ± 11.6	0.957
Gender, female, number (%)	64 (80.0)	44 (80.0)	20 (80.0)	1.000
Age of diagnosis, years, mean, ± SD	35.8 ± 11.9	36.0 ± 12.1	35.2 ± 11.8	0.798
Years of disease mean ± SD	6.8 ± 5.7	6.7 (5.9)	7.2 (5.5)	0.674
Comorbidities, number (%)				
Hypertension	23 (28.8)	15 (27.3)	8 (32.0)	0.791
Diabetes	5 (6.2)	3 (5.5)	3 (12.0)	0.370
Coronary artery disease	2 (2.5)	2 (3.6)	0 (0.0)	1.000
Dyslipidemia	6 (7.5)	3 (5.5)	3 (12.0)	0.370
Thromboembolic event	57 (71.2)	22 (88.0)	35 (63.6)	**0.033**
Cerebrovascular disease	30 (37.5)	18 (32.7)	6 (24.0)	0.599
Hashimato disease	7 (8.8)	6 (10.9)	1 (4.0)	0.425
Chronic kidney disease	7 (8.8)	4 (7.3)	3 (12.0)	0.671
Obesity	9 (11.2)	8 (14.5)	1 (4.0)	0.260
Others	8 (10.0)			
Primary APS, number (%)	15 (18.8)	13 (23.6)	2 (8.0)	0.128
Secondary APS, number (%)	65 (81.2)	42 (76.4)	23 (92.0)	0.128
Rheumatoid arthritis	1 (1.2)	1 (1.8)	0 (0.0)	
Lupus	57 (71.2)	38 (69.1)	19 (76.0)	
Undifferentiated connective tissue disorder	1 (1.2)	1 (1.8)	0 (0.0)	
Sjögren's syndrome	6 (7.5)	2 (3.6)	4 (16.0)	
Symptoms				
Fever	7 (8.8)	4 (7.3)	3 (12.0)	0.671
Weight loss	9 (11.2)	5 (9.1)	4 (16.0)	0.450
Fatigue	14 (17.5)	8 (14.5)	6 (24.0)	0.348
Livedo reticularis	6 (7.5)	6 (10.9)	0(0.0)	0.169
Raynaud	11 (13.8)	9 (16.4)	2 (8.0)	0.488
Amourosis fugax	8 (10.0)	7 (12.7)	1 (4.0)	0.424
Avascular necrosis	2 (2.5)	2 (3.6)	0 (0.0)	1.000
Arthritis	29 (36.2)	20 (36.4)	9 (36.0)	1.000
Nephritis	13 (16.2)	7 (12.7)	6 (24.0)	0.326
Laboratory findings				
Hemolytic anemia	7 (8.8)	6 (10.9)	1 (4.0)	1.000
Leukopenia	12 (15.0)	8 (14.5)	4 (16.0)	0.744
Thrombocytopenia	14 (17.5)	11(20.0)	3(12.0)	0.681
ANA positivity	73 (91.2)	50 (90.9)	23 (92.0)	1.000
Ds DNA positivity	39 (48.8)	27 (49.1)	12 (48.0)	1.000
Low complement	41 (51.2)	30 (54.5)	11 (44.0)	0.471
Coombs positivity	22 (27.5)	16 (29.1)	6 (24.0)	0.789
Antibodies included in APS classification criteria				
Anti-cardiolipin IgG	43 (53.8)	31 (56.4)	12 (48.0)	0.629
Anti-cardiolipin IgM	21 (26.2)	17 (30.9)	4 (16.0)	0.184
Anti beta 2 glycoprotein IgG	36 (45)	26 (47.3)	10 (40.0)	0.631
Anti beta 2 glycoprotein IgM	28 (35)	20 (36.4)	8 (32.0)	0.803
Lupus anticoagulant	55 (68.8)	33 (60.0)	22 (88.0)	0.018
APS associated non-criteria antibodies (n: number of patients tested)				
Anti phospholipid antibody IgG (n: 34)	12 (35.3)	0 (0.0)	12 (35.3)	1.000
Anti phospholipid antibody IgM (n:40)	11 (27.5)	0 (0.0)	11 (27.5)	1.000
Anti-cardiolipin IgA (n: 4)	0 (0.0)	0 (0.0)	0 (0.0)	

Table 1 (continued)

	N = 80	Non_criteria antibody tested (n:55)	Non criteria antibody untested (n:25)	p
Anti beta 2 glycoprotein IgA (n:35)	12 (34.3)	0 (0.0)	12 (34.3)	1.000
Anti phosphatidylserine antibody IgA (n:11)	0 (0.0)	0 (0.0)	0 (0.0)	
Anti phosphatidylserine antibody IgM (n:15)	2 (13.3)	0 (0.0)	2 (13.3)	1.000
Anti phosphatidylserine antibody IgG (n:14)	5 (35.7)	0 (0.0)	5 (35.7)	1.000
APS non-criteria antibody tested, number (%)	55 (68.8)			
APS non-criteria antibody positive, number (%)	29 (52.7)			
APS classification				
Obstetric APS	23(28.8)	20 (36.4)	3 (12.0)	**0.033**
Thrombotic APS	39 (48.8)	26 (47.3)	13 (52.0)	0.033
Obstetric + Thrombotic APS	18 (22.5)	9 (16.4)	9 (36.0)	0.033
Thrombotic APS	57 (71.2)	35 (63.6)	22 (88.0)	0.033
DVT	27 (33.8)	13 (23.6)	14 (56.0)	0.010
PTE	8 (10.0)	6 (10.9)	2 (8.0)	1.000
SVO	30 (37.5)	21 (38.2)	9 (36.0)	1.000
VCI thrombosis	3 (3.8)	2 (3.6)	1 (4.0)	1.000
Retinal vein thrombosis	2(2.5)	2 (3.6)	0 (0.0)	1.000
Portal vein thrombosis	1 (1.25)	1 (1.8)	0 (0.0)	1.000
Distribution of thrombosis according to patients				
DVT	19 (23.8)	7 (12.7)	12 (48.0)	
PTE	3 (3.8)	2 (3.6)	1 (4.0)	
SVE	23 (28.8)	17 (30.9)	6 (24.0)	
DVT + PTE	2 (2.5)	2 (3.6)	0 (0.0)	
DVT + PTE + SVE	2 (2.5)	2 (3.6)	0 (0.0)	
DVT + SVE	1 (1.2)	0 (0.0)	1 (4.0)	
DVT + VCI thrombosis + SVE	3 (3.8)	2 (3.6)	1 (4.0)	
Retinal vein thrombosis	2 (2.5)	2 (3.6)	0 (0.0)	
SVE + PTE	1 (1.2)	0 (0.0)	1 (4.0)	
Portal vein thrombosis	1 (1.2)	1 (1.8)	0 (0.0)	
Obstetric APS	41 (51.3)	29 (52.8)	12 (48.0)	

APS Antiphospholipid syndrome, *DVT* Deep Vein Thrombosis, *PTE* pulmonary thromboembolism, *SVE* Cerebrovascular Event, *VCI* Vena Cana Inferior

of patients can be aided by non-criteria aPLs [21]. The identification of novel aPLs has provided additional insights into the pathogenic mechanisms. While some aPLs have shown clinical or diagnostic value, the application of non-criteria aPLs is limited in clinical settings. The potential usefulness of evaluating non-criteria antibodies for predicting APS in patients is uncertain. The study faced challenges in assessing the clinical significance of non-criteria antibodies due to the limited number of cases and variability in the antibodies analyzed in each patient.

The literature includes many studies evaluating antibodies that do not conform to established criteria. Several investigations [22] have examined the diagnostic efficacy of aCL/aβ2GpI IgA in APS but resulted in inconsistent outcomes. Nevertheless, it is recommended by guidelines to conduct IgA testing in cases where aPL criteria continue to be negative [23]. It has been demonstrated that aCL IgA may be a potential risk factor for pregnancy morbidities in patients with APS [24]. Despite being tested in our clinic, aCL IgA has only been evaluated in a limited number of patients and yielded negative results in all cases. Consequently, the assessment of the association with the clinic was not feasible.

The investigation of aPS/PT prevalence in homogeneous aPLs revealed that patients who tested positive for LAC (84.5%) and triple positivity (83.4%) exhibited a higher pooled prevalence of aPS/PT IgG/M [24]. The diagnostic significance of aPS/PT IgG in APS has been validated in a multicenter study conducted on a global scale [25]. Research has shown that the presence of aPS/PT, especially with elevated levels of antibodies, is linked to the identification of thrombotic APS [26]. A prospective evaluation was conducted to determine the clinical significance of IgG/IgM aPS/PT antibodies in a cohort comprising 191 aPL carriers.

Table 2 Comparison of non-criteria aps antibody-positive patients with non-criteria aps antibody-negative patients

	Negative N = 26	Positive N = 29	p-value
Age, years, median (IQR)	41 (35,48)	41(32,55)	0.89
Gender, female, number (%)	25 (96.0)	19 (66.0)	0.005
Age of diagnosis, years, median (IQR)	32 (28,43)	35 (26,48)	0.64
Years of disease median, (IQR)	5 (3.2,10.8)	4 (2.0,8.0)	0.30
Comorbidities, number (%)			
Hypertension	4 (15.0)	11(38.0)	0.061
Diabetes	2 (7.7)	1 (3.4)	0.60
Coronary artery disease	1 (3.8)	1 (3.4)	> 0.99
Dyslipidemia	1 (3.8)	2 (6.9)	> 0.99
Cerebrovascular disease	7 (%27)	11 (%38)	0.39
Hashimato disease	5 (19.0)	1 (3.4)	0.090
Chronic kidney disease	2 (7.7)	2 (6.9)	> 0.99
Obesity	3 (12.0)	5 (17.0)	0.71
Primary APS, number (%)	9 (35.0)	4 (14.0)	
Secondary APS, number (%)	17 (65.0)	25 (86.0)	
Rheumatoid arthritis	0 (0.0)	1 (3.4)	
Lupus	15 (58.0)	23 (79.0)	
Undifferentiated connective tissue disorder	1 (3.8)	0 (0.0)	
Sjögren's syndrome	1 (3.8)	1 (3.4)	
APS classification			0.35
Obstetric APS	12 (46.0)	8 (28.0)	
Thrombotic APS	11 (42.0)	15 (52.0)	
Obstetric + Thrombotic APS	3 (12.0)	6 (21.0)	
Thrombosis area			0.38
DVT	2 (7.7)	5 (17.0)	
PTE	2 (7.7)	0 (0.0)	
SVE	6 (23.0)	11 (38.0)	
DVT + PTE	0 (0.0)	2 (6.9)	
DVT + PTE + SVE	1 (3.8)	1 (3.4)	
DVT + SVE	0 (0.0)	0 (0.0)	
DVT + VCI thrombosis + SVE	1 (3.8)	1 (3.4)	
Retinal vein thrombosis	1 (3.8)	1 (3.4)	
SVE + PTE	0 (0.0)	0 (0.0)	
Portal vein thrombosis	1 (3.8)	0 (0.0)	
VTE	8 (31.0)	10 (34.0)	0.77
Arterial Thrombosis	8 (31.0)	13 (45.0)	0.28
Laboratory			
Leukopenia	2 (7.7)	0 (0.0)	**0.016**
Thrombocytopenia	4 (15.0)	7 (24.0)	0.30
ANA positivity	24 (92.0)	26 (90.0)	> 0.99
Anti- ds DNA positivity	9 (35.0)	18 (62.0)	**0.042**
Low complement	10 (38.0)	20 (69.0)	**0.023**
Antibodies included in APS classification criteria			
Anti-cardiolipin IgG	8 (31.0)	23 (79.0)	**< 0.001**
Anti-cardiolipin IgM	7 (27.0)	10 (34.0)	0.54
Anti beta 2 glycoprotein IgG	5 (19.0)	21 (72.0)	**< 0.001**
Anti beta 2 glycoprotein IgM	9 (35.0)	11 (38.0)	0.80
Lupus anticoagulant	14 (54.0)	19 (66.0)	0.38

1 Median (IQR); n (%) 2 Wilcoxon rank sum test; Pearson's Chi-squared test; Fisher's exact test,

APS Antiphospholipid syndrome, *DVT* Deep Vein Thrombosis, *PTE* pulmonary thromboembolism, *SVE* Cerebrovascular Event, *VCI* Vena Cana Inferior

The study revealed that IgG PS/PT antibodies are associated with an increased risk of thrombosis [27]. The present investigation revealed a noteworthy IgM aPS/PT incidence among subjects exhibiting exclusive LAC positivity. The study revealed noteworthy correlations between venous thrombotic occurrences and IgG aPS/PT, as well as between pregnancy loss and IgG/IgM aPS/PT [15].

The research investigated the prospective utility of atypical aPLs in practice. The study assessed patients for the existence of various antibodies, including PS/PT and phosphatidic acid, phosphatidyl-ethanolamine, phosphatidyl-glycerol, phosphatidyl-inositol, phosphatidylserine, annexin V, prothrombin IgM, and IgG antibodies, as well as aβ2GP1 IgA and aβ2GP1 domain 1 IgG antibodies. The findings indicated a correlation between the severity of APS and the presence of aforementioned antibodies and that non-criteria aPLs may offer advantages to patients categorized as seronegative APS. Additionally, it suggests that anti-PS/PT antibodies could function as a substitute for LA in the classification of high-risk patients undergoing treatment with direct oral anticoagulants (DOACs) in cases where LA detection is not a viable option [13]. The assessment of these antibodies has predominantly been conducted on patients who exhibit a negative serological status. The investigation of clinical correlation has been conducted among seronegative patients. Contrary to the aforementioned research, the clinical implications of seropositivity in patients remain ambiguous.

Another research assessed a cohort of 175 individuals diagnosed with APS, 122 individuals diagnosed with other autoimmune diseases (as a control group for disease), and 50 healthy individuals to determine the presence of seven non-criteria aPLs. These included aPS/PT of IgG/IgA/IgM isotypes, anti-phosphatidylethanolamine (aPE) antibodies of IgG/IgA/IgM isotypes, anti-annexin V antibodies of IgG/IgA/IgM isotypes, and anti-phosphatidylserine (aPS) antibodies of IgM and IgG isotypes, as well as antibodies directed against a mixture of phospholipids (APhL) of IgG and IgM isotypes. The present investigation revealed that annexinV had the highest frequency of non-criteria antiphospholipid antibodies (58.86%). Furthermore, the inclusion of non-criteria antiphospholipid antibodies was demonstrated to enhance the precision of antiphospholipid syndrome diagnosis. Antiphospholipid IgG, aPS/PT, and anti-phosphatidyl serine IgG, APS, have been proposed as plausible biomarkers for anticipating thrombotic risk. Another research assessed a cohort of 312 individuals, comprising 100 patients who received a diagnosis of APS, 51 patients who had APS as a secondary condition to SLE, 71 patients with SLE, and 90 healthy controls. The study subjects underwent testing for aCL IgG/IgM/IgA, aβ2GPI IgG/IgM/IgA, aPS/PT IgG/IgM, and anti-annexin A5 antibodies (aAnxV) IgG/IgM. The findings of this investigation indicate that aCL IgA and aAnxV IgM have the potential to aid in the identification of seronegative APS patients, while aPS/PT IgG is linked to stroke [5].

The relationship between non-criteria antibodies and obstetric APS has garnered significant attention in recent years. A comparative investigation was conducted to analyze the clinical characteristics, laboratory data, and fetal-maternal outcomes of 640 women diagnosed with obstetric complications associated with aPL that did not satisfy Sydney criteria (non-criteria obstetric antiphospholipid syndrome, NC-OAPS) and 1000 women diagnosed with obstetric APS (OAPS). The results revealed significant variations in clinical and laboratory parameters between the two groups. Nonetheless, comparable fetal-maternal outcomes were noted in both cohorts after receiving treatment [28]. Also, the study revealed that individuals diagnosed with OAPS exhibited a greater prevalence of low birth weight, fetal loss, stillbirth, early-onset placental vascular pathology (PE < 34 weeks and FGR < 34 weeks), and prematurity in comparison to the NC-OAPS cohort. All patients diagnosed with OAPS in our study exhibited positivity for at least one antibody per the specified criteria. Currently, there is insufficient comprehensive research to evaluate the impact of criterion-unrelated antibody positivity on pregnancy-related mortality and morbidity among obstetric patients with APS.

APS is frequently observed in association with SLE, with a prevalence rate of around 35% [1]. ApLs are frequently detected in individuals with SLE, with a positivity rate ranging from 30 to 40% [29]. Common factors may contribute to the development of APS and SLE [30], and the generation of antiphospholipid antibodies may have a hereditary component [31]. The study aimed to assess the plausible pathogenic associations between SLE and APS development in patients with or without secondary APS. The study scrutinized "criteria" and "non-criteria" aPLs concerning SLE's biological parameters and secondary APS diagnosis. According to the research findings, there was a significant correlation between the IgG serotypes of "non-criteria" aPLs and the production of anti-DNA. In contrast, the IgM serotypes were linked to the consumption of complement C3 [29]. In our study, most patients exhibited secondary APS in conjunction with SLE. We also found a significant association between non-criteria antibody positivity and dsDNA positivity and complement deficiency. Thus, we formulated a hypothesis suggesting that the presence of non-criteria antibodies may be linked to the immunological activity of SLE. Furthermore, it has been observed that the frequency of aPL that does not meet the predetermined criteria is more prevalent in individuals with SLE and secondary APS when compared to those with SLE who lack APS characteristics. The presence of these aPL increases the risk of thrombotic events [32]. Furthermore, a study conducted in Poland examined the presence of aPE and aPS antibodies in individuals diagnosed with SLE. The results of the study indicated that patients who have aPS

and aPE antibodies are at risk of developing vascular complications. In particular, the presence of aPE significantly increases the likelihood of developing thrombotic complications in SLE patients who do not have classical serological markers of APS [33].

It should be noted that this investigation is subject to certain limitations. The primary and most significant limitation of our research is the restricted sample size. Enhancing the sample size and incorporating patients with a broader spectrum of associated conditions or clinical features could enhance the reliability of the results. A notable limitation was the absence of a control group. Additionally, the study includes patients with various clinical features. The lack of uniformity in evaluating non-criteria antibodies among patients has resulted in inconsistent assessments of identical antibodies across different patients.

Although there are many studies in the literature evaluating the contribution of non-criteria antibodies to the diagnosis and their relationship with clinical findings, this is the first to evaluate the relationship between non-criteria antibody positivity and concomitant autoimmune disease in secondary APS patients. Although non-criteria antibodies have no role in the diagnosis of APS, non-criteria antibody positivity may be associated with high immunological activity of the underlying disease. In patients with positive non-criteria antibodies, care should be taken in terms of underlying disease activation.

Conclusion

Limited research has been conducted to assess the comprehensive range of non-criteria autoantibodies within identical patient cohorts. Additional investigation is warranted to establish their associations with more intricate clinical manifestations. We found that non-criteria antibody positivity may be associated with high immunological activity of the underlying disease. The precise nature of the association between antibodies that satisfy the established criteria and those that do not remains unclear. Additionally, the role of non-criteria antibodies in the clinical manifestation of seropositive APS patients necessitates further inquiry. The clinical usefulness of non-criteria antibodies is limited due to the lack of standardized measurement techniques and restricted accessibility. There is a requirement for extensive, multi-institutional research to examine the medical importance of the presence of aPLs, either alone or in combination, with or without criteria.

Author contribution All the authors made a substantial contribution to this research. All members contributed to the study design, data collection, writing the paper, and approved the final form.

Data availability The data of this study are available on request from corresponding author.

Declarations

Conflict of interest The authors declare no competing interests.

References

1. Cervera R, Serrano R, Pons-Estel G et al (2015) Morbidity and mortality in the antiphospholipid syndrome during a 10-year period: a multicentre prospective study of 1000 patients. Ann Rheum Dis 74(6):1011–1018
2. Tektonidou MG, Andreoli L, Limper M et al (2019) EULAR recommendations for the management of antiphospholipid syndrome in adults. Ann Rheum Dis 78(10):1296–1304
3. Miyakis S, Lockshin MD, Atsumi T et al (2006) International consensus statement on an update of the classification criteria for definite antiphospholipid syndrome (APS). J Thromb Haemost 4(2):295–306
4. Hughes G, Khamashta M (2003) Seronegative antiphospholipid syndrome. Ann Rheum Dis 62(12):1127
5. Hu C, Li S, Xie Z et al (2021) Evaluation of the diagnostic value of non-criteria antibodies for antiphospholipid syndrome patients in a Chinese cohort. Front Immunol 12:741369
6. Zohoury N, Bertolaccini ML, Rodriguez-Garcia JL et al (2017) Closing the serological gap in the antiphospholipid syndrome: the value of "non-criteria" antiphospholipid antibodies. J Rheumatol 44(11):1597–1602
7. Sciascia S, Bertolaccini M (2014) Antibodies to phosphatidylserine/prothrombin complex and the antiphospholipid syndrome. Lupus 23(12):1309–1312
8. Mekinian A, Bourrienne M-C, Carbillon L et al (2016) Non-conventional antiphospholipid antibodies in patients with clinical obstetrical APS: prevalence and treatment efficacy in pregnancies. Seminars in arthritis and rheumatism. Elsevier
9. Liu X, Zhu L, Liu H et al (2022) Non-criteria antiphospholipid antibodies in antiphospholipid syndrome: diagnostic value added. Front Immunol 13:972012
10. Otomo K, Atsumi T, Amengual O et al (2012) Efficacy of the antiphospholipid score for the diagnosis of antiphospholipid syndrome and its predictive value for thrombotic events. Arthritis Rheum 64(2):504–512
11. Sciascia S, Sanna G, Murru V et al (2015) The global antiphospholipid syndrome score in primary APS. Rheumatology 54(1):134–138
12. Nakamura H, Oku K, Amengual O et al (2018) First-line, non-criterial antiphospholipid antibody testing for the diagnosis of antiphospholipid syndrome in clinical practice: a combination of anti–β2-glycoprotein I domain I and anti–phosphatidylserine/prothrombin complex antibodies tests. Arthritis Care Res 70(4):627–634
13. Litvinova E, Darnige L, Kirilovsky A et al (2018) Prevalence and significance of non-conventional antiphospholipid antibodies in patients with clinical APS criteria. Front Immunol 9:2971
14. Devreese KM, Zuily S, Meroni PL (2021) Role of antiphospholipid antibodies in the diagnosis of antiphospholipid syndrome. J Transl Autoimmun 4:100134
15. Shi H, Zheng H, Yin Y-F et al (2018) Antiphosphatidylserine/prothrombin antibodies (aPS/PT) as potential diagnostic markers and risk predictors of venous thrombosis and obstetric complications in antiphospholipid syndrome. Clin Chem Lab Med 56(4):614–624

16. Žigon P, Podovšovnik A, Ambrožič A et al (2019) Added value of non-criteria antiphospholipid antibodies for antiphospholipid syndrome: lessons learned from year-long routine measurements. Clin Rheumatol 38:371–378
17. Radin M, Foddai SG, Cecchi I et al (2020) Antiphosphatidylserine/prothrombin antibodies: an update on their association with clinical manifestations of antiphospholipid syndrome. Thromb Haemost 120(4):592–598
18. Yin D, Chayoua W, Kelchtermans H et al (2020) Detection of anti-domain I antibodies by chemiluminescence enables the identification of high-risk antiphospholipid syndrome patients: a multicenter multiplatform study. J Thromb Haemost 18(2):463–478
19. Ortona E, Capozzi A, Colasanti T et al (2010) Vimentin/cardiolipin complex as a new antigenic target of the antiphospholipid syndrome. Blood J Am Soc Hematol 116(16):2960–2967
20. Zuo Y, Willis R, Papalardo E et al (2017) A unique antiphospholipid assay recognizing phospholipid mixture compared with criteria antiphospholipid immunoassays in lupus patients. Lupus 26(6):606–615
21. Liu T, Gu J, Wan L et al (2020) "Non-criteria" antiphospholipid antibodies add value to antiphospholipid syndrome diagnoses in a large Chinese cohort. Arthritis Res Ther 22(1):1–11
22. Bertolaccini ML, Amengual O, Andreoli L et al (2014) 14th International Congress on Antiphospholipid Antibodies Task Force. Report on antiphospholipid syndrome laboratory diagnostics and trends. Autoimmun Rev 13(9):917–930
23. Lakos G, Favaloro EJ, Harris EN et al (2012) International consensus guidelines on anticardiolipin and anti–2-glycoprotein I testing. Arthritis Rheum 64(1):1–10
24. Zhu R, Cheng C-Y, Yang Y et al (2022) Prevalence of aPhosphatidylserine/prothrombin antibodies and association with antiphospholipid antibody profiles in patients with antiphospholipid syndrome: a systematic review and meta-analysis. Thromb Res 214:106–114
25. Amengual O, Forastiero R, Sugiura-Ogasawara M et al (2017) Evaluation of phosphatidylserine-dependent antiprothrombin antibody testing for the diagnosis of antiphospholipid syndrome: results of an international multicentre study. Lupus 26(3):266–276
26. Vandevelde A, Chayoua W, de Laat B et al (2022) Added value of antiphosphatidylserine/prothrombin antibodies in the workup of thrombotic antiphospholipid syndrome: communication from the ISTH SSC Subcommittee on Lupus Anticoagulant/Antiphospholipid Antibodies. J Thromb Haemost 20(9):2136–2150
27. Tonello M, Mattia E, Favaro M et al (2019) IgG phosphatidylserine/prothrombin antibodies as a risk factor of thrombosis in antiphospholipid antibody carriers. Thromb Res 177:157–160
28. Alijotas-Reig J, Esteve-Valverde E, Ferrer-Oliveras R et al (2020) Comparative study of obstetric antiphospholipid syndrome (OAPS) and non-criteria obstetric APS (NC-OAPS): report of 1640 cases from the EUROAPS registry. Rheumatology 59(6):1306–1314
29. Dima A, Caraiola S, Jurcut C et al (2015) Extended antiphospholipid antibodies screening in systemic lupus erythematosus patients. Rom J Intern Med 53(4):321–328
30. Sestak A, O'Neil K (2007) Familial lupus and antiphospholipid syndrome. Lupus 16(8):556–563
31. Radway-Bright E, Ravirajan C, Isenberg D (2000) The prevalence of antibodies to anionic phospholipids in patients with the primary antiphospholipid syndrome, systemic lupus erythematosus and their relatives and spouses. Rheumatology 39(4):427–431
32. Szodoray P, Tarr T, Tumpek J et al (2009) Identification of rare anti-phospholipid/protein co-factor autoantibodies in patients with systemic lupus erythematosus. Autoimmunity 42(6):497–506
33. Fischer K, Przepiera-Będzak H, Brzosko I et al (2022) Anti-phosphatidylethanolamine and anti-phosphatidylserine antibodies—association with renal involvement, atherosclerosis, cardiovascular manifestations, Raynaud phenomenon and disease activity in Polish patients with systemic lupus erythematosus. Biomolecules 12(10):1328

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