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IgA anti-β2 glycoprotein I antibodies: Experience from a large center

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ABSTRACT

Objective: IgG and IgM antibodies directed at β2-glycoprotein I are included in the classification criteria for the antiphospholipid syndrome (APS) while the IgA antibodies against β2-glycoprotein I (IgA aβ2GPI) are not. Conflicting data about the significance of IgA aβ2GPI and APS manifestation can be found and more studies are necessary in order to define the diagnostic value of IgA aβ2GPI. In the present article, we investigated the possible role of IgA aβ2GPI as marker of APS.

Methods: A cohort of 314 patients with APS and systemic autoimmune disease was investigated for the presence of IgA aβ2GPI and its association with clinical manifestation of APS.

Results: Eighty-nine patients presented IgA aβ2GPI, 68 cases associated with others antiphospholipid antibodies (aPL) and in 21 cases being the only aPL present. In primary APS IgA aβ2GPI are highly coincidental with other aPL (92,2%) while most of the isolated IgA aβ2GPI were present in the SLE group (16/21).

No association between IgA aβ2GPI and APS manifestations: thrombosis and pregnancy morbidity was found, while a positive association between IgA aβ2GPI and the presence of anti-nDNA, anti-RNP, anti-Sm, anti-SSA, anti-SSB antibodies was encountered.

Conclusion: Our study does not show association between IgA aβ2GPI and APS manifestations and does not support the inclusion of IgA aβ2GPI as a classification criteria for APS.

1. Introduction

The Antiphospholipid Syndrome (APS), a common autoimmune disease, clinically manifests as recurrent thrombosis and/or pregnancy-related morbidity accompanied by the presence of antiphospholipid antibodies (aPL). In order to fully qualify as APS, 1 clinical and 1 laboratory classification criteria need to be met [1]. The laboratory classification criteria include IgG/IgM antibodies directed to β2-glycoprotein I (β2GPI), either isolated (IgG/IgM aβ2GPI), or bound to cardiolipin (IgG/IgM aCL), together with the presence of Lupus Anticoagulant (LA).

A number of other (auto)antibodies, still considered as non-classification APS criteria, are currently under study worldwide: IgA anti-β2GPI antibodies (IgA aβ2GPI), anti-β2GPI domain I antibodies (anti-DI Ab), anti-phosphatidylserine/prothrombin antibodies (aPS/PT), anti-phosphatidylethanolamine (aPE), Annexin A5 resistance assay [2].

Since 2012, the IgA isotype of the aPL (IgA aβ2GPI and IgA aCL) has been accepted as a laboratory classification criteria for Systemic Lupus Erythemathosus (SLE) [3]. The presence of IgA aβ2GPI has been linked to atherosclerotic disease, macrovascular disease in scleroderma patients and cardiovascular mortality in hemodialysis patients [4,5,6]. When looking to the clinical features of APS (arterial and venous thrombosis and pregnancy morbidity) one may find studies both favoring [7,8,9,10,11,12,13] as well as denying [14,15] a role for IgA aβ2GPI in APS pathogenesis. The 14th International Congress on Antiphospholipid Antibodies Task Force revising the available data on IgA aPL although finds that IgA aβ2GPI present a more consistent association with APS manifestations than IgA aCL assigns a grade III level of

Abbreviations: aβ2GPI, anti-beta2 glycoprotein I antibodies; aCL, anti-cardiolipin antibodies; ANA, anti-nuclear antibodies; aPS/PT, antiphosphatidylserine/prothrombin antibodies; APS, Anti-phospholipid Syndrome; AT, arterial thrombosis; β2GPI, beta2 glycoprotein I; C3/4, complement factor C3/4; IgA/G/M, immunoglobulin A/G/M; LA, lupus anticoagulant; MTCD, Mixed Tissue Connective Disease; PM, pregnancy morbidity; SBI, silent brain isquemia; silent aPL, presence of anti-phospholipid antibodies without clinical manifestation of APS; SLE, Systemic Lupus Erithematosus; SS, Sjögren Syndrome; TH, thrombosis; VT, venous thrombosis

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evidence - low quality of evidence for IgA aPL [16]. This view is still supported by recent reviews of non-criteria aPL and their correlation with APS manifestations [17,18]

The contradictory results of the clinical significance of IgA aβ2GPI stimulated the present investigation. The aim of this study was to evaluate the clinical associations of IgA aβ2GPI in our patient population, in an effort to better understand their diagnostic utility.

2. Patients and methods

Inclusion criteria: 314 consecutive patients, ≥ 18 years old, referred between 1997 and 2012 to the Systemic Autoimmune Diseases Unit of Hospital La Paz, Madrid, Spain, with suspicion of APS or related autoimmune diseases. All patients gave informed consent and the study was approved by the Ethics Board of Hospital La Paz. A complete medical history, physical examination, and complementary studies were systematically performed in all the patients at inclusion. We collected data on: age, gender, thrombotic arterial events (stroke, transient ischemic attack (TIA), myocardial infarction (MI), silent brain ischemia, other arterial thrombotic events), venous thrombosis (deep vein thrombosis (DVT), pulmonary embolism (PE), other venous thrombotic events) and pregnancy morbidity [abortion, fetal deaths, premature births, intrauterine growth retardation, pre-eclampsia, eclampsia, HELLP syndrome (hemolysis, elevated liver enzymes, low platelets counts)]. Features associated with APS were also recorded: heart valve disease, thrombocytopenia, livedo reticularis, migraine, and nephropathy. The presence of other pro-thrombotic risk factors was investigated: smoking habit, arterial hypertension, hiperlipemia, traumatisms, diseases (diabetes, cancer, nephrotic syndrome), obesity, immobility, medication (corticotherapy, oral contraceptives), antithrombin deficiency, C and S protein deficiency, factor V Leyden, G20210A prothrombin mutation, high levels of homocysteine.

After the initial visit, the patients were reviewed every 6 months, or sooner, if required by the patient's clinical condition. The data were collected in a Microsoft Access database using 2 types of standardized forms: an inclusion form, summarizing the patient's medical record, and a follow-up form, containing patient's evolution and laboratory data between 2 consecutive visits. Serum samples, collected prospectively, were stored at − 20 °C. APS related manifestations were evaluated and considered as indicated by the current APS classificatory criteria [1] and only criteria aPL were used for diagnostic classification. When the diagnostic process was completed, the patients were classified following two characteristics: thrombosis (primary APS, secondary APS, silent presence of aPL, no aPL) or autoimmune disease (for example: Systemic Lupus Erythematosus - SLE, Sjögren Syndrome - SS, Mixed Tissue Connective Disease - MTCD or no autoimmune disease). Infection was investigated and excluded both at inclusion and during the follow-up. Briefly, 167 patients were classified as presenting APS (116 primary APS, 51 secondary APS), 63 patients presented aPL without related clinical manifestation, 83 patients did not presented the classical aPL or aPS/PT, 125 patients presented SLE, 17 SS and 3 patients presented MTCD. All the patients were treated according to the guidelines available at the time of their enrolment. APS patients were treated either with antiaggregant therapy (mainly aspirin) or with anticoagulant therapy (acenocoumarol) with a targeted International Normalized Ratio (INR) between 2.7 and 3.5. During pregnancy, acenocoumarol therapy was substituted for low-molecular-weight heparin (LMWH). Most patients with SLE received hydroxychloroquine and nonsteroidal anti-inflammatory drugs (NSAIDS). Acute flares were treated with steroids and other immunosuppressant drugs, as necessary. A detailed description of the clinical parameters that were collected is published in our previous work [19], as well as the methods employed for the detection of classical aPL (IgG/IgM aCL, IgG/IgM aβ2GPI, LA) and for anti-phosphatidylserine/prothrombin antibodies (IgG/IgM aPS/PT). From the samples utilized for the quantification of the classificatory aPL, one sample per patient was tested for IgA aβ2GPI presence using Inova Diagnostics QUANTA Lite® β_2 GPI IgA ELISA kit (ref 708675) following the kit's manufacturer instructions. The cut-off for positivity was established with sera from 100 healthy controls, and a value higher than 20 U/ml was considered positive. The selection criterion for the samples to be tested for IgA aβ2GPI presence was proximity to the diagnostic moment. When such a sample could not be used, a sample which best reflected the patients aPL profile over time was tested.

The presence of Antinuclear Antibodies (ANA) was determined by Indirect Immunefluorescence (IFI) on Hep-2 slides (Euroimmun). A panel of anti-DNAn antibodies and Extractable Nuclear Antigens (SSA, SSB, RNP, Sm, etc.) was performed by multiplex assay (ANA AtheNA Multi-Lyte). Renal function was assessed by measuring creatinine levels, 24 h proteinuria and the presence of hematuria, renal casts and leukocyturia. MDRD4-IDMS was calculated using the following formula: $GFR = 175 \times SerumCr^{-1.154} * age^{-0.203} * 1.212$ (if the patient is african-american) * 0.742 (if female) [20]. For statistical analysis, the creatinine result which best reflected the time trend of the renal function was chosen.

2.1. Statistical analysis

Data were collected using a Microsoft Access database, and statistical analysis was performed with SPSS v11.5. Quantitative variables were described using average, standard deviation, mean and range. Qualitative variables were described using absolute and relative frequencies expressed in percentages. Continuous quantitative variables were compared mainly by non-parametric tests: Kruskal-Wallis or U of Mann-Whitney and a $p < 0,05$ was considered as significative. Qualitative variables were compared by Fisher's exact test. The degree of correlation between quantitative variables was calculated by Pearson correlation coefficient. Multivariate analysis was performed in order to identify the independent factors that are positively associated with the dependent variable and to control for possible confounding parameters; multivariate logistic regression model were designed. The magnitude of each factor was expressed as an odds ratio (OR). A stepwise model was followed, computing only the variables associated with the dependent variable in a previous univariate analysis. The model's goodness of fit was expressed as an area under receiver operating characteristics (ROCs) curve and 95% confidence interval (95% CI). Results were grouped in tables showing the final step of the variables together with the corresponding b coefficient, standard error (SE), statistical significance, and eb term, which represents the magnitude of effect adjusted by the rest of the variables and expressed as OR.

3. Results

3.1. Cut-off for IgA aβ2GPI assay

The adequacy of the cut-off recommended by the manufacturer was tested using sera from 100 healthy volunteers following the APS classification criteria recommendations [1,21]. The results ranged from 1.13 U/ml to 32.73 U/ml, (mean 6.6 U/ml) and only 2 sera gave results over 20 U/ml. Our calculated cut-off for the 99th percentile was found to be 20.72 U/ml, close to the one appointed by the manufacturer. Therefore, the proposed cut-off of 20 U/ml was accepted.

3.2. Prevalence of IgA aβ2GPI antibodies

A total of 314 patients were investigated for the presence of IgA aβ2GPI, of whom 89 (39,55%) were positive for the presence of IgA aβ2GPI with values included in the range of 20.6 U/ml - 296 U/ml, (mean: 68.3 U/ml, SD 63.02 U/ml). In 21 patients, IgA aβ2GPI was the only detectable aPL (Table 1). Table 2 shows the IgA aβ2GPI prevalence coincident with other aPL. The presence of IgA aβ2GPI correlated with the presence of most aPL except IgM aCL. The highest rate of

Table 1
Presence of IgA a2βGPI in the study group.

(n)	IgA aβ2GPI + (n, %)
Patients (314)	89 (28,34%), 21 times isolated
Healthy controls (100)	2 (2%)
Diagnostic	
APS (167)	51 (30.53%)
Primary APS (116)	37 (31.9%)
Secondary APS (51)	14 (27.45%)
Silent aPL (63)	20 (31.74%)
No aPL (83)	18 (21.68%)
SLE (125)	38 (30.4%)
SS (17)	8 (47.05%)
MTCD (3)	0 (0%)
Manifestation	
AT (87)	27 (31%)
Myocardial Infarction (15)	6 (40%)
Stroke (37)	12 (32.4%)
TIA (31)	10 (32.25%)
AT Retina (7)	3 (42.85%)
AT Renal (2)	0 (0%)
Silent brain isquemia (50)	15 (30%)
VT (61)	22 (36.06%)
DVT (52)	17 (32.7%)
PE (19)	8 (42.1%)
Renal vein thrombosis (1)	1 (100%)
Pregnancy morbidity (74)	20 (27%)

Table 2
IgA aβ2GPI prevalence, compared to other aPL, in the study group.

IgA aβ2GPI relationship with positivity for other aPL				
aPL (n)			IgA aβ2GPI + (n, % aPL, *p*)	
aCL	IgG	110	45 (40,9%)	< 0.001
	IgM	110	35 (31,8%)	0.316
aβ2GPI	IgG	64	34 (53,12%)	< 0.001
	IgM	57	23 (40,35%)	0.021
aPS/PT	IgG	60	28 (46,7%)	< 0.001
	Ig	66	25 (37,9%)	0.041
AL		131	54 (41,22%)	< 0.001

coincidence was with the IgG isotype (aβ2GPI, aPS/PT, aCL).

3.3. Clinical association of IgA aβ2GPI

Considering the entire cohort, IgA aβ2GPI showed no association with the main clinical classification criteria for APS: arterial thrombosis group (AT), venous thrombosis group (VT) or pregnancy morbidity group (PM) (Table 3a). It was neither associated with any specific type of thrombosis: stroke, acute myocardial infarction, deep vein thrombosis, pulmonary embolism, silent brain ischemia.

As expected from the data above, IgA aβ2GPI showed no correlation with APS diagnosis (Table 3b), opposed to the rest of the aPL studied who did show association with APS diagnosis and its clinical manifestations.

Next, we separately investigated the performance of IgA aβ2GPI within the non-SLE group and non-primary APS group, following previous work of Despierres et al. [8] who found that different groups of patients (SLE vs. non-SLE) present IgA a2βGPI that target different domains (domain I versus domain IV/V) of β2GPI.

3.4. Clinical association of IgA aβ2GPI Ab in the non-SLE group

After excluding the 125 patients presenting SLE, 189 patients were analyzed (115 primary APS, 12 secondary APS, 62 other diagnostics). IgA aβ2GPI was present in 51 patients, 5 times isolated and 46 (92,2%) times coincided with others aPL. IgA aβ2GPI was found to be positively associated with venous thrombosis ($p = 0.028$, OR 2.375 95%CI: 1.145–4.928). No association was encountered between arterial thrombosis or pregnancy morbidity and IgA aβ2GPI presence. IgA aβ2GPI positivity and APS diagnostic almost reached statistical significance ($p = 0.064$, OR 2.033 95%CI: 1.008–4.098) as did presenting low levels of complement C4 ($p = 0.054$, OR 2.544 95%CI: 1.064–6.083). No other parameter associated with IgA aβ2GPI, including decreased renal function.

Due to the very high coincidence of IgA aβ2GPI with other aPL, further analysis of the strength of association between IgA aβ2GPI and VT was performed. ROC curve and a step-wise multivariate logistic regression model for the presence of TV (including IgG/IgM aCL, IgG/IgM aβ2GPI, IgG/IgM aPS/PT, LA) were constructed. IgA aβ2GPI ROC curve area for VT was found to be 0.556 (95%CI: 0.473–0.640) (Table 4). The regression model excluded the IgA aβ2GPI as an independent factor associated with VT, showing that the best predictive capacity for VT it is displayed by the triple positivity for IgG aCL, IgG aPS/PT y AL with a ROC curve area of 0.778 (95%CI: 0.697–0.859).

3.5. Clinical association of IgA aβ2GPI Ab in the non-primary APS group

After excluding the 115 patients presenting primary APS, 199 patients were analyzed (125 patients presenting SLE and 51 secondary APS). IgA aβ2GPI was positive in 52 patients, 16 cases isolated, and 36 times (69.2%) associated with the presence of others aPL. IgA aβ2GPI showed no association with the main clinical classification criteria for APS: arterial thrombosis, venous thrombosis, pregnancy morbidity. Also no association was found with the SLE diagnostic, a positive association was calculated with the presence of anti-nDNA ($p = 0.018$ OR 2.564 95%CI: 1.226–5.362), anti-RNP ($p = 0.017$, OR 3.009 95%CI: 1.250–7.239) and anti-Sm antibodies ($p = 0.009$, OR 4.236 95%CI: 1.477–12.150). The association between IgA aβ2GPI and anti-Sm and anti-RNP antibodies maintained when the analysis was performed within the entire group of 314 patients.

Complement consumption was not associated with the presence of IgA aβ2GPI. When renal function was investigated, the presence of IgA

Table 3a
Lack of IgA aβ2GPI association, compared to other aPL, with major APS manifestations.

		AT			VT			PM		
		p	OR	CI95%	*p*	OR	CI95%	*p*	OR	CI95%
aβ2GPI	IgA	ns (0.49)	1.2	0.71–2.06	ns (0.155)	1.56	0.86–2.83	ns (0.123)	0.57	0.28–1.14
aCL	IgG	**0.008**	1.95	1.20–3.16	**0.001**	4.07	2.32–7.13	ns	1.28	0.68–2.40
	IgM	**0.008**	1.94	1.20–3.16	ns	0.87	0.49–1.54	ns	0.95	0.49–1.82
aβ2GPI	IgG	ns	1.6	0.92–2.77	**0.001**	3.92	2.18–7.05	0.065	2.05	0.96–4.38
	IgM	ns	1.62	0.9–2.92	ns	1.52	0.78–2.94	ns	1.59	0.69–3.64
aPS/PT	IgG	ns	0.9	0.5–1.6	**0.001**	7.64	4.12–13.93	**0.047**	2.36	1.04–5.39
	IgM	ns	1.06	0.6–1.9	**0.027**	2.07	1.13–3.78	ns	1.37	0.64–2.91
AL		ns	1.45	0.9–2.33	**0.001**	8.2	4.25–15.80	ns	1.25	0.67–2.34

Table 3b
IgA aβ2GPI association, in comparison with other aPL, with APS and SLE diagnostic.

Fisher's exact test (p)	IgA aβ2GPI	IgG aCL	IgM aCL	IgG aβ2GPI	IgM aβ2GPI	IgG aPS/PT	IgM aPS/PT	AL
APS	0.455	**< 0.001**	**< 0.001**	**< 0.001**	**< 0.001**	**< 0.001**	**< 0.001**	**< 0.001**
SLE	0.611	**0.005**	**< 0.001**	**0.014**	**< 0.001**	0.683	0.075	**< 0.001**

In bold: aPL with positive association with APS and SLE.

Table 4
ROC curve area for Venous Thrombosis in the non-SLE group.

		Area	95% CI
aβ2GPI	IgA	0.556	0.473–0.64
aCL	IgG	0.676	0.594–0.758
	IgM	0.493	0.41–0.576
aβ2GPI	IgG	0.617	0.534–0.701
	IgM	0.524	0.449–0.597
aPS/PT	IgG	0.685	0.603–0.767
	IgM	0.605	0.523–0.678
AL		0.754	0.685–0.823

aβ2GPI was not associated with high levels of creatinine or low levels of glomerular filtration rate (MDRD4-IDMS < 60 ml/min/1.73m^2).

3.6. Analysis of isolated presence of IgA aβ2GPI

As stated above, 21 patients presented IgA aβ2GPI as the only detectable aPL (IgG/IgM aCL, aβ2GPI, aPS/PT and LA negative). If the presence of LA was admitted, the number of patients IgA aβ2GPI-only would ascend to 23. When this 2 groups of patients (21 and 23 respectively) were compared to the patients having no aPL at all (73 patients aCL IgG/IgM negative, aβ2GPI IgA/IgG/IgM negative, aPS/PT IgG/IgM negative, LA negative), no positive correlation could be established between IgA aβ2GPI presence and the main clinical classification criteria for APS (Table 5). A synthetic description of the 5 women with pregnancy morbidity and isolated presence of IgA aβ2GPI is depicted in Table 6.

However, a positive correlation for isolated IgA aβ2GPI was found with the presence of the Sjögren Syndrome, anti-SSA, anti-SSB, anti-Sm and anti-RNP antibodies. Again, decreased renal function was not associated with the presence of IgA aβ2GPI.

Within the 125 patients with SLE, the highest number of patients (16) presenting isolated IgA aβ2GPI was found. Within this group the only positive association of isolated IgA aβ2GPI was the presence of anti-Sm and anti-RNP antibodies. No association was found with thrombosis, pregnancy morbidity or decreased renal function.

4. Discussion

With the present investigation we studied the prevalence, clinical and paraclinical associations of IgA aβ2GPI in serum samples of a cohort of 314 consecutive patients with APS or systemic autoimmune diseases.

From the 314 patients, IgA aβ2GPI was present in 89 cases (28.34%), generally associated with another aPL. Only 21 patients (6.68%) presented isolated IgA aβ2GPI with the majority of cases appearing in SLE patients (16/21, 76.2%). We could not establish a positive correlation between the presence of IgA aβ2GPI and the clinical manifestation of APS (arterial thrombosis, venous thrombosis, pregnancy morbidity).

The same finding remains true when isolated or independent IgA aβ2GPI was analyzed. When a "flexible" definition of isolated IgA aβ2GPI, (admitting LA positivity) was applied, the number of patients who were considered positive for IgA aβ2GPI ascended from 21 to 23. Such modest increase, suggesting a poor association between isolated IgA aβ2GPI and LA is consistent with the poor clinical performance described above. Therefore, the high coincidence found between LA and IgA aβ2GPI in our cohort is explained by the concomitant presence of other aPL such as IgG aCL, IgG/M β2GPI or IgG/M aPS/PT, not by an intrinsic LA producing activity of IgA aβ2GPI.

In our cohort, 5 women with a history of pregnancy morbidity presented IgA aβ2GPI as the only aPL detectable, but a positive association between these 2 parameters could not be found. In 3 of the 5 cases, the pregnancy events, probably related to their autoimmune disease (2 SLE, 1 SS), were followed by normal deliveries of a healthy baby despite not receiving antiaggregant or anticoagulant therapy. This observation, despite being relative (the serum samples tested were posterior to the pregnancy events), further doubts a pathogenic role for IgA aβ2GPI in PM.

Our results are in line with the recent findings of Tebo et al. [15], where isolated IgA aβ2GPI fails to associate with APS clinical manifestation in the Hopkins Lupus cohort. However, in the smaller PROMISSE cohort, a kit dependent positive association for IgA aβ2GPI was obtained for pre-eclampsia (Inova kit) and miscarriage (Phadia kit). Again, no association was found between thrombosis and isolated IgA aβ2GPI in the PROMISSE cohort.

Opposed to our conclusions, the group of Martinez-Flores et al. recently found that circulating immune complexes of isolated IgA aβ2GPI and β2GPI (B2A-CIC) are associated with the occurrence of acute thrombotic events [22]. However, it is not clear if B2A-CIC are the cause of acute thrombosis or if they are produced once the thrombotic event is initiated.

In the last decade, sub-lytic activation of the complement cascade has emerged as a pivotal event in creating a pro-thrombotic environment, a mechanism shared with variable intensity by various entities: paroxysmal nocturnal hemoglobinuria, atypical haemolytic uremic syndrome, APS [23]. We know now that β2GPI is a regulator of the alternative complement pathway and probably of the classical pathway too [24]. There is evidence that in APS the classical pathways is activated [25,26]. Anti-β2GPI antibodies, especially anti-domain I antibodies, probably initiate complement activation by 2 mechanisms: 1) inhibition of complement regulation and 2) classical pathway activation through the constant domains of the antibodies in case of IgG1, IgG3, IgM isotypes. The IgA isotype weakly (if at all) activates complement [27]. Therefore IgA aβ2GPI probably have a lower influence on the complement pathway than the IgG/IgM counterparts and thus a lower pro-thrombotic activity.

It is true that complement activation seem not to be the only

Table 5
Description of the patients who presented isolated IgA aβ2GPI.

Isolated IgA aβ2GPI: IgG/IgM aCL/aβ2GPI/aPS/PT negative LA negative			
IgA aβ2GPI + (n = 21)		IgA aβ2GPI- (n = 73)	p[b]
AT	3 (1 MI, 2 TIA)[a]	7	0.688
SBI	1	13	0.18
VT	1	4	1
SLE	16	47	0.229
SS	4	3	0.042
IgA aβ2GPI + (n = 16)		IgA aβ2GPI- (n = 41)	p[b]
PM	5	21	0.24

[a] These patients also presented other risk factors for arterial thrombosis.
[b] Fischer's exact test p value for the IgA aβ2GPI + group.

Table 6
Description of the 5 patients with isolated IgA aβ2GPI and Pregnancy morbility.

Patient	Diagnostic	Pregnancy manifestation	Normal pregnancies	Treatment	
				Antiaggregant or anticoagulant	Other
1	SS	Fetal loss (16w)	Yes (before and after)	No	Hydroxychloroquine
2	SLE	IGR	Yes (after)	No	Hydroxychloroquine
3	SLE	Fetal loss (12w)	Yes (after)	No	No
4	SLE	2 abortions (< 10w)	Yes (before)	No	Hydroxychloroquine
5[a]	Diabetes	Fetal loss (12w)	yes (after)	No	Antidiabetics

IGR: intrauterine growth retardation.
[a] Important presence of other pro-thrombotic risk factors.

mechanism by which aPL mediate their effect: an important number of receptors were discovered for aβ2GPI/β2GPI immune complexes (Toll-like receptors -TLR 2, 4, 8, annexin A2, LDL-receptor family proteins) [28], their effect being mediated via the phosphatidylinositol 3-kinase (PI3K)–RAC serine/threonine-protein kinase (AKT) pathway - mechanistic target of rapamycin (mTOR) or P38 mitogen-activated protein kinase (p38 MAPK) [29,30]. Recently it was shown that IgG anti-domain I β2GPI promotes apoptosis via p38 MAPK in a rat model [31]. Conversely, domain V of β2GPI protects from apoptosis-induced cell death [32].

Interestingly, the presence of IgA aβ2GPI was associated in our results with the presence of anti-Sm and anti-RNP antibodies. This finding is in line with the inclusion of IgA aPLs as a classification criteria for LES [3]. Moreover, isolated IgA aβ2GPI was also associated with anti-SSA and anti-SSB antibodies. Despite the fact that these antibodies contribute to the general antibody burden found in SLE and SS, they do not seem to be linked to APS manifestation. Maybe they delineate a group of patients with higher morbidity as found by Serrano et al. and Delgado et al. in different patient settings [6,33].

The positive association between IgA aβ2GPI and anti-Sm antibodies suggested the study of renal function in our cohort of patients, but no correlation between decreased renal function and IgA aβ2GPI could be established. This finding might be explained by the fact that all the patients classified as suffering from systemic autoimmune disease received treatment.

In conclusion, our study does not find a significant association between IgA aβ2GPI and APS manifestations. Isolated IgA aβ2GPI tend to appear in a non-thrombotic SLE setting rather than in a APS one and show a low prevalence. Therefore, larger cohorts or meta-analisis are needed in order to define their role in APS.

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Disclosure statement

The authors declare no relevant conflict of interest regarding this article.

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