Anticardiolipin Antibodies in Juvenile Rheumatoid Arthritis and Systemic Lupus Erythematosus

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ABSTRACT

Background: Antiphospholipid antibody syndrome (APS) can either occur as a primary syndrome or associated with other autoimmune diseases such as systemic lupus erythematosus (SLE). Anticardiolipin antibody (aCL) of IgG and/or IgM isotype in blood, measured by a standardized ELISA is the most acceptable laboratory criteria. APS IgG isotype, particularly IgG2 subclass is more strongly associated with thrombosis. **Objectives:** This study was done to determine the prevalence of IgG aCL and its subclasses in relation to APS symptoms, in a group of juvenile rheumatoid arthritis (JRA) and juvenile systemic lupus erythematosus (SLE) patients. **Methods:** In this prospective study, 28 JRA and 16 SLE patients, aged 3-18 years, were enrolled. IgG aCL was assayed by standard aCL ELISA. IgG subclasses were also assayed by ELISA on sera with medium to high titers of aCL. ACL assay was performed on at least two occasions for each patient, over 3-6 months period of follow up. **Results:** 29% (8/28) of JRA patients and 44% (7/16) of SLE patients had aCL. Six of SLE patients displayed APS related manifestations: hemolytic anemia, thrombocytopenia, arterial occlusion, valvular heart disease, livedo reticularis and pulmonary hypertension, but none of them had persistant medium or high titer of aCL. The lack of association of high titer of aCL with APS related symptoms was observed in two patients. The IgG subclasses were primarily IgG1 and IgG3. **Conclusion:** The prevalence of IgG aCL in this group of pediatric SLE and JRA is not uncommon but it's relation to clinical manifestations is not clear. IgG1 and IgG3 subclasses were not associated with thrombosis, which is in agreement with previous studies.

Key words: Antiphospholipid Syndrome, Anticardiolipin Antibody, Juvenile Rheumatoid Arthritis, Systemic Lupus Erythematosus

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INTRODUCTION

Antiphospholipid antibodies (aPL) have been a subject of great interest since the beginning of the last century first because they were found to be serodiagnostic markers for syphilis and later because of their association with thrombosis and pregnancy loss in an autoimmune syndrome called the antiphospholipid antibody syndrome (APS) (1). It can either occur as a free-standing syndrome (primary APS) or associated with other autoimmune diseases, such as systemic lupus erythematosus (SLE) (secondary APS) (2). aPL consists of two groups of autoantibodies so called lupus anticoagulant (LA) and anticardiolipin antibodies (aCL) (3).

Most of the information available on aPL-associated clinical features come from studies carried out in adults. Data concerning pediatric patients drive from various case reports and a few studies on a larger number of patients (4).

The aims of this study were to investigate the prevalence of aCL, its subclasses and their correlation with related clinical events in a cohort of juvenile rheumatoid arthritis (JRA) and pediatric SLE patients.

PATIENTS AND METHODS

Patients. Two groups of patients were enrolled in this prospective study. Patients with JRA and patients with SLE who were followed at Motahari Out Patient Clinic or admitted at Nemazee Hospital Immunology Ward, Shiraz, Iran, during one year period (1999-2000).

JRA patients. 28 patients were studied, their age at the time of study ranged from 3 to 18 years. Fourteen patients were female, and fourteen were male.

The diagnosis was based on the American college of Rheumatology (ACR) criteria (5) . Ten patients had systemic JRA, eleven had pauciarticular and seven had polyarticular JRA (table 1).

SLE patients. Sixteen patients were studied. Their age at time of this study ranged from 8-19 years. Fourteen patients were female and 2 were male. SLE diagnosis was based on the ACR criteria (6). ANA was negative in the last three patients, despite the presence of at least three clinical criteria (table 2).

Methods. Detection of IgG aCL was detected by an enzyme linked immunoassay (ELISA) method, prepared according to the method of Gharavi et al. (7), for two occasions in 3-6 months interval.

Calculation of the results as GPL (IgG antiphospholipid antibody) units, that can be used for standardization, was done by comparison with standard sera which were available as a kind gift of Dr Gharavi. This standard ELISA was set up at Nemazee Hospital Pediatric Immunology Research Lab. The titers of 10<20 GPLU, 20-80 GPLU and >80 GPLU were defined as low, medium and high titers, respectively (7).

IgG subclasses were determined by a modified ELISA procedure by using the mouse

Table 1. aCL in the JRA patients

Patient	Age (yr)	Sex	Onset type	ACL (GPL) First assay	ACL (GPL) Second assay
1	12	Male	Pauci	<10	<10
2	13	Male	Poly	<10	<10
3	14	Female	Sys	20.0	<10
4	5	Female	Pauci	<10	<10
5	14	Female	Sys	<10	<10
6	14	Male	Sys	<10	<10
7	14	Female	Poly	<10	<10
8	14	Female	Pauci	<10	<10
9	10	Female	Pauci	<10	<10
10	12	Male	Pauci	<10	<10
11	15	Male	Poly	<10	<10
12	18	Female	Sys	<10	<10
13	10	Male	Sys	<10	<10
14	5	Female	Pauci	42.0	<10
15	15	Male	Sys	25	17
16	13	Male	Pauci	<10	-
17	12	Female	Sys	<10	12.5
18	10	Male	Pauci	<10	-
19	3	Male	Sys	<10	-
20	5	Female	Sys	24	-
21	13	Male	Pauci	<10	-
22	7	Female	Sys	> 80	-
23	9	Male	Poly	<10	-
24	12	Female	Poly	14	-
25	8	Male	Pauci	<10	<10
26	9	Female	Poly	<10	-
27	12	Female	Poly	14	-
28	15	Male	Pauci	<10	-

monoclonal antibodies to human IgG subclasses.

Statistical analyses were done by chi-square test using SPSS software.

RESULTS

JRA patients. aCL were detected in 8/28 (29%) of patients, of which one had high titer (>80 GPLU), four had medium titer (20-80 GPLU), and three had low titer aCL. None had any symptoms related to APS (8). aCL was present mostly in patients with systemic JRA (table 1).

SLE patients. aCL were detected in 7/16 (44%) of patients of which, two had high titer and two had medium titer antibody. APS related symptoms are shown at Table 2. There was not any significant difference (P = 0.3), in prevalence of aCL between the two groups of enrolled patients.

IgG aCL subclasses. Detection of IgG aCL subclasses was performed on 9 sera with medium to high titer of aCL. IgG aCL was primarily IgG1 and IgG3, in this group of patients.

DISCUSSION

Definite antiphospholipid antibody syndrome is considered to be present if at least one of the clinical criteria (vascular thrombosis or pregnancy morbidity) and one of the laboratory criteria are met (9). Anticardiolipin antibody of IgG and/or IgM isotype in blood, present in medium or high titer, on two or more occasions, at least 6 weeks apart, measured by a standardized ELISA for anticardiolipin antibodies is the most acceptable laboratory-based criteria (3). There are some other clinical and laboratory features associated in some studies of aCL or APS, such as thrombocytopenia, hemolytic anemia, livedo reticularis, pulmonary hypertension, migraine, anti-2 glycoprotein I antibodies and low- positive titers of IgG, which show weaker associations than the mentioned definite criteria (9).

The number of enrolled patients and their distribution of age and sex in our study is similar to other studies (4). 8/28 (29%) of our JRA patients had aCL IgG, which is consistent with the previous reported prevalence from the same pediatric studies (4,10,11). 7/16 (44%) of pediatric SLE patients were aCL positive in this study. The reported prevalence of aCL antibodies in childhood SLE ranges from 30 to 87% (4,10,11). There was no APS related clinical feature among the JRA group which was expected (4,5,6). Although there are some reported cases of aCL positive JRA with thrombosis (12), in a more recent study included a large group of JRA patients, despite a high prevalence of aPL, there was no association between the aCL, anti-beta2 GP or lupus anticoagulant with clinical manifestations (13).

In 50% of systemic JRA patients aCL was found. We had also a case of systemic JRA with high titer of aCL for one occasion (table 1). Because of small number of patients, the comparison of aCL positive sera, among three onset types was not statisticaly possible.

5/16 lupus patients manifested with APS related symptoms in this study (table 2).

Patient no. 2, presented with hemolytic anemia at the onset of her disease which progressed to a complete criteria of lupus within 6 months, with high ANA, thrombocytopenia, malar rash and kidney disease. She developed the symptoms of non-acute arterial insufficiency a few months later which led to diagnosis of right subclavian artery occlusion. aCL was below 10 GPLU in her serum during our study period. Arterial occlusion may be multifactorial in this patient and we did not have a tissue evidence of the thrombotic nature of this arterial lesion. Patients 7 and 10 manifested with previous thrombocytopenia and heart valvular insufficiency associated with transient high and medium titers of aCL.

Patient no. 14 was an unfortunate girl with chronic arthritis and anemia, treated as a possible case of JRA who developed sudden severe autoimmune (Coomb's positive)

Table 2. aCL in the SLE patients

Patient	Age (yr)	Sex	ACL (GPL) First assay	ACL (GPL) Second assay	APS related clinical feature
1	13	Female	<10	<10	Previous hemolytic anemia and thrombocytopenia
2	14	Female	<10	<10	Arterial occlusion, previous hemolytic anemia and thrombocytopenia
3	14	Female	<10	<10	
4	14	Female	<10	<10	
5	14	Female	<10	<10	
6	18	Female	100.0	<10	
7	19	Female	85.0	<10	Previous thrombocytopenia
8	11	Male	<10	<10	
9	15	Female	Low+	32	
10	16	Male	36	<10	MVP+ MR
11	11	Female	Low+	12	
12	14	Female	<10	<10	
13	12	Female	<10	<10	Levido reticularis
14	11	Female	Low+	16.5	Hemolytic anemia + mild PH
15	8	Female	<10	--	
16	11	Female	19	<10	

hemolytic anemia. She did not respond to high doses of steroid, IVIG and plasmapheresis and passed away. She could be diagnosed as a probable case of APS with persistent low titer of aCL and negative ANA at time of hemolytic event. It is concluded in the previous studies that there is no association between the lupus activity and presence of aCL antibodies (4), and APS is a non inflammatory autoimmune disease (7).

One of the remarkable findings of our study is the lack of association of high titer of aCL and related clinical symptoms in our SLE patients, It means that none of our SLE patients met the criteria for definite APS.

In a similar study from Italy, 2 out of 18 SLE patients had a definite and one had a probable APS (4). The reasons for this disimilar finding are not easily apparent and may reflect differences in patients' ages or selection, ethnic background or even methodology used for aCL assays. In the other hand, 65% of Iranian adult patients with SLE expressed aCL in a recent study by Sammangooei et al. (14) with more prevalence of symptoms which may reflect the effect of age in the prevalence and pathogenecity of aCL.

In a more recent study, no association was found between aPL antibodies and clinical manifestations in a group of 57 children and adolescents with SLE, although clinical manifestation of APS was detected in eight patients (15).

IgG subclasses assay showed that in all of our 9 patients with medium to high titer of aCL, the predominant IgG subclasses were IgG1 and IgG3. Pathogenicity of aCL may be related to IgG2, associated with venous and arterial thrombosis (7). This finding is in accordance with the lack of related clinical symptoms in patients with high aCL in this study.

We conclude that IgG aCL in our pediatric SLE and JRA patients is common but there is no clear clinical association.

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