Letters to the Editors

Anti-annexin 5 in patients with systemic lupus erythematosus

Sirs,

Annexin-5 is a human ubiquitous phospholipid-binding protein that has affinity for anionic phospholipids, particularly phosphatidilserine, a component of plasma cell membrane (1). Exposure of phosphatidilserine in the cell outer membrane is a physiological signal for the onset of coagulation and apoptosis processes (1). Annexin-5 recognises this signal, binds to phosphatidilserine forming a shield that prevents an excessive phospholipid-dependent coagulation reaction and the pro-inflammatory activities of the dying cell (1).

Systemic lupus erythematosus (SLE) is an autoimmune disease in which the clearance of apoptotic cells is defective (2). IgM and IgG anti-annexin-5 (aANX-5) antibodies have been found in SLE patients and may interfere with annexin-5 function (3). It has been suggested that they mediate reduction in annexin-5 anticoagulant properties leading to thrombosis and/or vascular occlusion (4). Some authors have linked it to peripheral thrombosis and foetal losses (3); others have shown that they do not change pregnancy outcome (5). Hrycek *et al*. (6) have described that SLE patients on immunosuppression have higher levels of annexin-5 and IgG aANX-5 antibodies than those without it, suggesting that they are more common in patients with more severe disease.

In the present study, we analysed IgM and IgG aANX-5 in 89 consecutive non-pregnant female patients (convenience sample) with SLE, trying to relate them to clinical and serological expression of the disease and to disease activity measured by SLEDAI. This study was approved by the local Ethics Research Committee and all patients signed consent. Serum levels of IgG and IgM aANX-5 were measured by Elisa (Orgentec®) and the cut-off was set at 9U/ml. Charts were reviewed for cumulative presence of clinical signs, demographic and autoantibody data. Clinical signs were classified according to ACR-1997 SLE Classification Criteria (7). Antiphospholipid syndrome (APS) was diagnosed according to modified Sapporo Classification Criteria (8) and the non-criteria APS manifestations considered were migraine, pseudovasculitic skin ulcers, livedo reticularis and involuntary movement disorders (9). Disease activity index was determined by SLEDAI (10) concurrently with blood withdrawal for aANX-5 determination.

All patients were female with a mean age of 38.9±11.9 years and a median disease duration of 87 months. Is this population 69.7% had photosensitivity, 55.6% arthritis; 46.5% aphtae; 28.0% leucopenia; 24.8% nephritis; 13.8% serositis; 12.6% seizures; 10.3% discoid lesions; 8.9% haemolysis; 7.9% pschosis; 35.2% anti-Ro; 29.2% anti-dsDNA; 24.3% anti-RNP; 17.4% anti-Sm; 27.2% anticardiolipin IgG; 25.8% anticardiolipin IgM; 21.3% lupus anticoagulant and 19.7% anti-β2 GPI. In this population 20.2% had diagnosis of APS (9.6% with obstetrical criteria and 15.9% with vascular criteria and 4.4% with both). Anti ANX-5 IgG was positive in 17/89 (19.1%) and aANX-5 IgM in 4/89 (4.4%). All patients with aANX-5 IgM were also positive for aANX-5 IgG.

Table I. Clinical and serological profile of 89 patients with systemic lupus erythematosus according to the presence of anti-annexin 5 IgG.

	With anti-annexin 5 IgG n=17	Without anti-annexin 5 IgG n=72	*p*-value
Mean age (years)	38.2± 9.6	39.1±12.5	0.68*
Median disease duration (months)	111.0 (IQR 67.5 – 128.0)	87 (IQR 60.0 – 135.0)	0.52**
Arthritis	10/16 – 62.5%	39/72 – 54.1%	0.54§
Discoid lesions	2/17 – 11.7%	7/70 – 10%	1.00§§
Photosensitivity	10/17 – 58.5%	50/69 – 72.4%	0.27§
Butterfly rash	8/17 – 47.0%	32/72 – 44.4%	0.85§
Oral ulcer	12/17 – 70.5%	29/71 – 40.8%	0.03§
Psychosis	2/17 – 11.7%	5/71 – 7.0%	0.61§§
Seizures	0/17 – 0%	11/70 – 15.7%	0.11§§
Serositis	5/17 – 29.4%	7/72 – 9.7%	0.047§§
Nephritis	5/17 – 29.4%	26/72 – 36.1%	0.77§§
Leucopenia	4/17 – 23.5%	21/72 – 29.1%	0.76§§
Thrombocytopenia	5/17 – 29.4%	18/72 – 29.1%	0.76§§
Lymphocytopenia	3/17 – 17.6%	9/65 – 13.8%	0.70§§
Haemolysis	1/17 – 5.8%	7/72 – 9.7%	1.00§§
Anti dsDNA	3/17 – 17.6%	23/72 – 31.9%	0.37§§
Anti Ro	3/17 – 17.6%	28/71 – 39.4%	0.06§§
Anti La	0/17 – 0	17/71 – 23.9%	0.035§§
Anti Sm	3/17 – 17.6%	12/69 – 17.3%	1.00§§
Anti RNP	4/14 – 28.5%	16/68 – 23.5%	0.73§§
Anticardiolipín IgG	7/17 – 41.1%	17/71 – 23.9%	0.22§
Anticardiolipin IgM	8/17 – 47.0%	15/72 – 20.8%	0.026§
Lupus anticoagulant	9/17 – 52.9%	10/72 – 13.8%	0.0004§
Anti B2 GPI	6/17 – 35.2%	11/69 – 15.9%	1.00§§
Anti Annexin-5 IgM	4/17 – 23.5%	0/72 – 0%	0.001§§
Obstetrical APS	2/14 – 14.2%	6/69 – 8.6%	1.00§§
Vascular APS	6/17 – 35.2%	8/72 – 11.1%	0.023§
Total APS	6/17 – 35.2%	12/72 – 16.6%	0.10§

IQR: interquartile range; APS: Antiphospholipid syndrome.
* = unpaired *t*-test; ** = Mann Whitney test; § = chi squared test; §§ = Fisher test.

The results of our study of this population for association of aANX-5 IgG with clinical and serological findings through univariate analysis are presented in Table I.

When all variables with $p \leq 0.05$ (oral ulcers, serositis, anti La, aCl Ig M, aANX-5 IgM and lupus anticoagulant) were studied through logistic regression, only lupus anticoagulant remain significantly associated (OR=6.0; 95% CI=1.83-19.6; p=0.0031).

The SLEDAI varied from 0 to 16; patients with positive aANX-5 IgG had median value of 2 while in those without it was 0.0 (p=0.035).

We could not prove any association of aANX-5 with obstetrical, vascular APS neither with non-criteria APS manifestations . The possible association of aANX-5 with other antiphospholipid antibodies may obscure this relationship; in the present study a strong relationship with LA was found. Another interesting link was found between SLEDAI and positivity for aANX-5. It may be that this antibody may define patients with a more active disease.

In conclusion, our results show that patients with higher SLEDAI have higher positivity to aANX-5. Although we could not prove associations with thrombosis or foetal loss, patients with aANX-5 have more LA.

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