

## The Role of Anticardiolipin and Anti-B2 Glycoprotein Antibodies in Clinical Complication of Lupus Nephritis

Nawar Abass Abud Noor , Khalida Mossa Mousawy \*, Mohammed Abud Al akoo -Faham \*\*, Nehla Ganim and Leen Moustafa \*\*\*

### ABSTRACT:

#### BACKGROUND:

Although there was confliction regarding the association of raised anticardiolipin antibodies (ACL) level with renal disease in systemic lupus erythematosus (SLE), the role of B2glycoprotein as a cofactor for ACL binding is established. The presence of ACL & anti-B2GPI may be directly involved in pathogenesis of antiphospholipid antibodies associated symptoms like recurrent fetal loss, thrombocytopenia and thrombosis.

#### AIM OF STUDY:

To study the possible association between some auto antibodies with the most common clinical complication of disease.

#### PATIENTS AND METHODS:

The study was conducted on 25 patients with lupus nephritis, attended the renal clinic in specialized surgical hospital/ medical city /Baghdad, 25 lupus patients without nephritis and 25 healthy controls. Enzyme linked immunsorbant assay was used for detection for ACL, anti-B2GP.

#### RESULT:

Although there was no significant difference in mean concentrations of ACL and anti- B2GP between lupus nephritis and without nephritis ( $P>0.05$ ), lupus nephritis patients were more likely positive for ACL. Positivity of 100% For ACL & anti- B2GP were detected in thrombotic complication, in fetal loss complication patients more likely to be positive for ACL (75%), anti-B2GP (50%), in thrombocytopenia positivity of ACL & anti-B2GP (75%).

#### CONCLUSION:

There was no association between anti-B2GP and renal disease and presence of both ACL & anti-B2GP carry higher risk for thrombosis and recurrent fetal loss.

**KEY WORD:** anticardiolipin antibodies, anti-B2glycoprotein antibodies, lupus nephritis.

### INTRODUCTION:

Systemic lupus erythematosus (SLE) is an autoimmune □ multisystemic disease of protean manifestations and variable behavior <sup>(1)</sup>. Clinically it is remitting and relapsing disease of acute or insidious onset that may involve virtually any organ. Lupus nephritis is a serious disease whose prognosis can usually be improved dramatically by treatment, but for which the treatment is potentially toxic, prolonged, complex and difficult to plan and carry out <sup>(2)</sup>. Reports of the prevalence of antiphospholipid antibodies (APL) in SLE are myriad and have varied widely, depending on the antigen source and method used. In fact, studies have demonstrated that anywhere from 5% to 70% of lupus patients

produce some type of APL <sup>(3)</sup>. The clinical symptoms associated with anticardiolipin antibodies (ACL) include, but are not limited to, arterial and venous thrombosis, recurrent fetal loss, hemolytic anemia, and APL- associated nephropathy <sup>(4,5)</sup>. Similarly, binding of anti-B2glycoprotein antibodies to endothelial cell lead to it's activation and promote coagulation <sup>(6)</sup>. Beside, the usual function of B2GP as natural anticoagulant protein <sup>(7)</sup>.

In this study we try to shed light on the possible association of these autoantibodies with clinical complication of disease.

#### PATIENTS:

The study was conducted on 25 patients with lupus nephritis , 25 patients without nephritis as patient control, some admitted to Baghdad hospital and other attend the renal clinic in specialized surgical hospital/ medical city, they fulfilled four or more of the ACR criteria for classification of SLE and compared with 25 healthy control.

\*Department of Immunology & Microbiology/ College of Medicine/ Baghdad University.

\*\* Department Microbiology/ College of Medicine/ Baghdad University.

\*\*\* Department of Immunology/ Teaching Laboratories.

## COMPLICATION OF LUPUS NEPHRITIS

### METHODS:

Enzyme linked immunosorbant assay (ELISA) was used for detection of ACL and anti- B2GPI.

Microplates are coated with highly purified ACL, B2GP antigen respectively, and the principles of the tests based on formation of complex and enzymatic color reaction.

1-anti-ACL ELISA kit ( Biomaghreb Kit Ref.80515s).

2-anti-B2GP ELISA kit (BINDAZYME Kit Ref: k991801).

### RESULTS:

In this study, the mean concentration of ACL antibodies in lupus nephritis ( $19.02 \pm 18.07$ ) was not significantly different from that of patient without nephritis ( $23.59 \pm 21.02$ ). when comparing mean concentration of ACL antibodies in lupus nephritis and in those without nephritis with healthy control, mean of ACL antibodies in patient groups were significant more than that in healthy control ( $5.4 \pm 1.7$ ). Although, the mean concentration of anti-B2GP IgM in lupus without nephritis ( $18.76 \pm 16.32$ ) was high significantly more than its concentration in healthy control ( $6.12 \pm 1.59$ ), but anti-B2GP IgM

concentration in lupus nephritis ( $12.21 \pm 1.9$ ) was not significantly different from that in healthy control, also this true when comparing anti-B2GP IgM between lupus nephritis and without nephritis patients. Regarding anti-B2GP IgG, there were no significant differences in its mean concentrations among the studied groups at all as seen in table (1).

The number of patients positive for ACL or anti-B2GP antibodies with these complications was often too low for meaningful statistical evaluation. For thrombotic complication, the patients with this complication have the same positivity for ACL and anti-B2GP in both groups (100%). In thrombocytopenic complication the positive ACL and anti- B2GP antibodies were more in non nephritis lupus; in each group the positive ACL patients were equal to positive anti-B2GP patients. In fetal loss complication, the patients with this complication were more in lupus nephritis, and more likely to be positive for ACL (75%) than without nephritis patients (58.5%). Positivity for anti-B2GP was the same in both groups (50%).

Table (1) Comparison of mean of Auto-Antibodies level among studied groups.

Studied groups	No	Anti-cardiolipin Ab. (ACL)		Anti-β2 glycoprotein Ab. (IgM)		Anti-β2 glycoprotein Ab. (IgG)	
		Mean±SD	C.S	Mean±SD	C.S	Mean±SD	C.S
Lupus nephritis	25	$19.02 \pm 18.07$	0.02 S	$12.21 \pm 11.97$	0.132 NS	$13.8 \pm 12.72$	0.204 NS
Non nephritis lupus	25	$23.59 \pm 21.02$	0.002 HS	$18.76 \pm 16.32$	0.002 HS	$11.71 \pm 10.75$	0.421 NS
Healthy control	25	$5.4 \pm 1.708$	-	$6.12 \pm 1.509$	-	$8.12 \pm 3.28$	-
Total	75						
Lupus nephritis Vs without nephritis		0.426 NS		0.106 NS		0.639 NS	

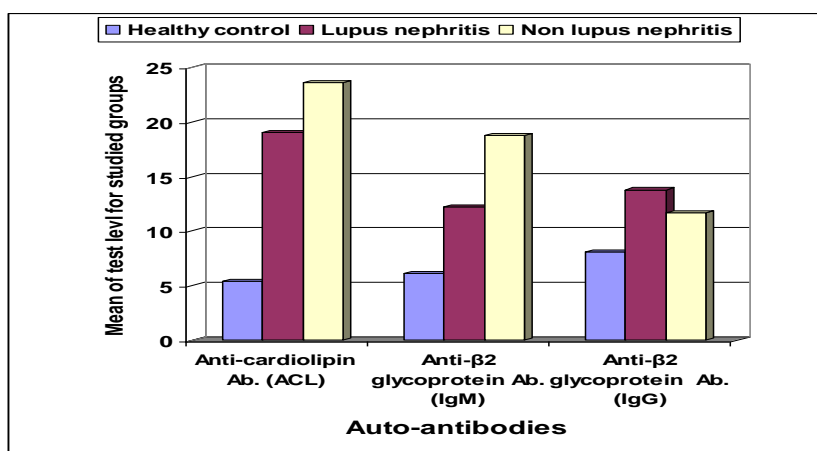


Figure (1) autoantibodies in studied groups

**Table (2) Distribution of patients according to Immunological tests.**

Immunological Tests			Lupus nephritis	lupus without nephritis	Total	Comparison of Significant
Anti-cardiolipin Ab. (ACL)	Positive	N.(%)	15 (60)	14 (56)	29 (58)	NS (P>0.05)
	Negative	N.(%)	10 (40)	11 (44)	21 (42)	
Anti-β2 glycoprotein Ab. (IgM)	Positive	N.(%)	10 (40)	11 (44)	21 (42)	NS (P>0.05)
	Negative	N.(%)	15 (60)	14 (56)	29 (58)	
Anti-β2 glycoprotein Ab. (IgG)	Positive	N.(%)	4 (16)	6 (24)	10 (20)	NS (P>0.05)
	Negative	N.(%)	21 (84)	19 (76)	40 (80)	

**Table (3): Comparison of the prevalence of Anti-cardiolipin Ab.(ACL) & Anti- β2 glycoprotein (Anti-B2GPI) with clinical complications in SLE Patients with & without nephritis.**

Patient group & Antibodies positivity	Thrombotic complication		Thrombocytopenia		Fetal loss	
	No.	%	No.	%	No.	%
Non nephritis- Lupus (n)	3		5		12	
ACL (+ve)	3	100	4	80	7	58.3
B2 GPI (+ve)	3	100	4	80	6	50
Lupus nephritis (n)	1		8		8	
ACL (+ve)	1	100	6	75	6	75
B2 GPI (+ve)	1	100	6	75	4	50
Healthy control (n)	0		0		0	
ACL (+ve)	0	0	0	0	0	0
B2 GPI (+ve)	0	0	0	0	0	0

**DISCUSSION:**

However, results of previous studies have been conflicting regarding the association of raised ACL level with renal disease in SLE patients; one study reported associations between ACL IgG and SLE nephritis<sup>(8)</sup>. In contrast other study found an inverse correlation between ACL and SLE nephritis<sup>(9)</sup>, while some reported no association at all<sup>(10)</sup>, which met this study. This difference could be due to difference in methods used and patient's selections. The ELISA technique was used in this study contain B2GP as a cofactor in plates, whereas the earlier studies used blocking solution contain little or no B2GP in their ACL assay, so this study measure B2GP dependent ACL while other studies measure B2PG independent ACL<sup>(11)</sup>. The finding for anti-B2GP antibodies was in agreement with those reported in previous studies that also failed to find an association between anti-B2GP and renal disease in SLE<sup>(12,13)</sup>, the presence of ACL and anti-B2GP in thrombotic complication this agree with many studies abroad<sup>(14, 15,16)</sup> which denote that raised levels of circulating antiphospholipid antibodies have been strongly associated with arterial, venous thrombosis, thrombocytopenia

and recurrent fetal loss in SLE and recently thrombosis has also been associated with raised levels of anti-B2GPI antibodies in both those studied patient groups<sup>(13)</sup>, because B2GP is anticoagulant factor in vivo. In thrombocytopenic complication, the percentage of occurrence of positive ACL and anti-B2GP IgG, IgM in lupus nephritis was 75% for each antibody which was less than that of lupus without nephritis 80%. This disagree with loizou, who denote that patients with thrombocytopenia in lupus nephritis group were more likely to be positive for ACL or B2GP than non nephritis lupus<sup>(17)</sup>, this may be due to frequent use of cytotoxic drugs by lupus nephritis patient in this study, which lead to decrease level of autoantibody and pancytopenia. For fetal loss complication, results agree with fact that multiple factors have been identified in association with less successful outcome in pregnancy include: lupus activity, previous nephropathy, maternal hypertension and positivity for APL<sup>(18, 19)</sup>. It also agrees with Di Simone who report that binding of anti-B2GP antibody to trophoblast down-regulates trophoblast human chorionic gonadotrophin

synthesis and secretion. Such a mechanism might contribute to defective placentation in women with fetal loss<sup>(20)</sup>.

## CONCLUSION:

This study demonstrates there were no association between anti-B2GP and renal disease and presence of both ACL and B2GP carry higher risk for thrombosis and recurrent fetal loss.

## REFERENCES:

1. Bevrá Hanneh Hahn 1960-1967. Principles of Internal Medicine by Herrisson. 16<sup>th</sup> Edition. MC Graw-Hill. Italia. Vol. 5.
2. Cameron JS. 1997. Systemic lupus erythematosus In: Immunological renal disease, edited by Nielson EG, Cousero WG, Philadelphia, Lippincott-Raven, pp 1055-1094.
3. Cervera R., Piette JC, Font J., Khamashta MA, Shoenfeld Y., Camps MT. et al. 2002. For the Euro-phospholipid Project Group. Anti-phospholipid syndrome: clinical and immunological manifestations and patterns of disease expression in a cohort of 1000 patients. *Arth. Rheum.* ,46:1019-1027.
4. Drenkard C., Villa AR, Alarcon-Segovia D., Porez-Vasquez MF. 1994. Influence of the antiphospholipid syndrome in the survival of patients with SLE. *J. Rheumatol.*,21:1067-72.
5. Gulko PS, Reveille JD, Koopman WJ, Burgard SL, Bartolucci AA, Alarcon GS. 1993. Anticardiolipin antibodies in SLE: Clinical correlation, HLA associations, and compact on survival. *J. Rheumatol.*,20:168-93.
6. Nawroth PP and Stern DM. 1987. Endothelial cell procoagulant properties and the host response. *Semin Thromb.*,13:391-7.
7. Kandiah DA, Krilis SA. 1994. B2GP-lupus, 3: 207-12.
8. Mc Hugh NJ, Maymo J, Skinner RP, et al. 1988. Anticardiolipin antibodies, livedo reticularis and renal disease in SLE. *Arth. Rheum. Dis.*;28:2108-14.
9. Weidman CE, Wallace DJ, Peter JB, et al. 1988. Studies of IgG, IgM and IgA antiphospholipid antibody isotypes in SLE. *Rheum.*,15:75-81
10. Frampton G, Hick J, Cameron JS. 1991. Significance of antiphospholipid antibody in patients with lupus nephritis. *KI*;39:1225-31.
11. Matsuda J., Saitoh N., Gohchi K., et al. 1993. Beta2 glycoprotein I-dependent and independent anticardiolipin antibody in patients with end-stage renal disease. *throm. res.* ;72:109-17.
12. Tsutsumi A., Matsuura E., Ichikawa K., et al. 1996. Antibody to B2GP and clinical manifestation in patient with SLE. *Arth. Rheum.*,39:1466-74.
13. Tsutsumi A., Matsuura E., Ichikawa K., Fujisaku A., Mukai M., Koike T. 1998. IgA class anti-B2GP in patients with SLE. *J. Rheum.* 25:74-8.
14. Harris EN, Ghan JKH, Asherson RA, et al. 1986. Thrombosis, recurrent Fetal loss, thrombocytopenia: predictive value of the anticardiolipin antibody test. *Arch Inter. Med.*,146:2153-6.
15. Loizou S., Byron MA, Englert HJ, et al. 1988. Association of quantitative anticardiolipin antibody levels with fetal loss and time of loss in SLE. *Q. J. Med.*,68:525-31.
16. Love PL and Sontoro SA. 1990. Antiphospholipid antibodies: anticardiolipin and lupus anticoagulant in SLE and in non-SLE disorders. *Ann. Rheumatol. Med.*, 112: 682-48.
17. Loizou S, Samarkos M., Norsworthy PJ, et al. 2000. Significance of anticardiolipin and anti-B2GP antibodies in lupus nephritis. *Rheumatology.*, 39:962-68.
18. Lima F., Buchanan N., Khamashta M., et al. 1994. Gynecological and Obstetric outcome in SLE, *Semin. Arth. Rheum.*, 25:184-92.
19. Petri M. 1994. SLE and pregnancy. *Rheum. Dis. Clin. North. AM*,20:67-118.
20. Di Simone, Raschi E., Castellani R. et al. 2005. Pathologic role of anti-B2GP antibodies in antiphospholipid syndrome associated fetal loss; characterization of anti-B2GP binding to trophoblast cells and functional effects of anti-B2GP antibodies in vitro. *Ann. Rheum. Dis.* 64:462-67.