Original Article

Prevalence of Lupus Anticoagulant in Women with Spontaneous Abortion in Zaria

IN Ibrahim, AI Mamman, SE Adaji[1], A Hassan, AA Babadoko

Departments of Haematology and [1]Obstetrics and Gynaecology, Ahmadu Bello University Teaching Hospital, Zaria, Kaduna, Nigeria

Abstract

Introduction: Spontaneous abortion (SA) is a common complication of pregnancy. Presence of lupus anticoagulant (LA), one of the antiphospholipid antibodies, has been associated with SA in many studies, especially in Caucasians. This study was carried out to determine the prevalence of LA in women with SA in ABUTH, Zaria. **Materials and Methods:** A cohort of 100 consecutive women presenting with SA with no history of thrombotic episodes were enrolled into the study. Prothrombin time (PT), kaolin clotting time (KCT), and activated partial thromboplastin time (APTT) were conducted on samples of all the participants. Eight patients had prolonged APTT, and after a 50:50 mixture of their plasma with pooled control plasma, four (50%) had uncorrected APTT. Staclot® (a hexagonal-phase phospholipid) test and calculated Rosner index for prolonged KCT were used for the confirmation of LA in samples with uncorrected APTT after mixing studies. **Results:** We analyzed 100 women with one or more SA with a mean age of 31.0 ± 3.8 years. Nearly 4% and 3% of the participants were LA positive with Staclot® and KCT tests, respectively. Patients with LA were more likely to have had a past history of preeclampsia/eclampsia, small for gestational age deliveries, and previous SA (prevalence odds ratio [95% confidence interval]) of 1.9 (0.2, 20.1), 3.2 (0.3, 34.3), and 1.4 (0.1–13.6), respectively. The PT, APTT, and KCT were significantly prolonged in patients with LA ($P \leq 0.001$ for each, respectively). **Conclusion:** LA may be one of the causes of SA and other adverse pregnancy outcomes such as preeclampsia/eclampsia and small for date deliveries. It is recommended that patients with prolonged APTT, uncorrected with 50:50 mixing study with pooled control plasma, should be evaluated further for LA.

Keywords: *Lupus anticoagulant, spontaneous abortion, uncorrected activated partial thromboplastin time*

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Introduction

Lupus anticoagulant (LA) is one of the antiphospholipid (APL) autoantibodies, which prolong phospholipid-dependent coagulation reactions that depend on protein-phospholipid complexes *in vitro*.[1,2] APL antibodies are a mix of several immunoglobulins (Ig): IgG, IgM, and less commonly, IgA antibodies directed at phospholipid-associated proteins, particularly prothrombin and beta 2-glycoprotein 1 (β2-GP1).[3] LA is a common cause of prolongation of phospholipid-dependent clotting assays such as activated partial thromboplastin time (APTT) and the dilute Russell's viper venom time.[4] Paradoxically, LA has been related to thrombotic events,[5] but could lead to bleeding in the presence of thrombocytopenia and other coagulopathy.[5,6]

Although LA was originally described in patients with systemic lupus erythematosus (SLE), it was detected subsequently in non-SLE disorders as well as in healthy individuals.[7] Positivity for anti-mitochondrial

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Address for correspondence: Dr. IN Ibrahim, Department of Haematology, Ahmadu Bello University Teaching Hospital, P. M. B. 06 Shika, Zaria - 810 001, Kaduna, Nigeria.
E-mail: dribronda@yahoo.com

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type 5 antibodies (anti-M5) had been demonstrated in a patient with inflammatory hepatobiliary and kidney changes in a background of APL syndrome.[8] Although anti-M5 has been regarded as a distinct serological marker for APL syndrome, the demonstration that β2-GPI can bind to anionic phospholipids and overall that it can react with mitochondrial particles supports the argument that it could be related to anti-β2-GPI activity.[9]

Positive results in tests for APL antibodies are associated with adverse pregnancy outcomes such recurrent pregnancy losses, or one or more late-term (>10 weeks' gestation) spontaneous abortions (SAs).[10] Other pregnancy complications include intra-uterine growth retardation, maternal thrombosis, thrombocytopenia, and pregnancy-induced hypertension.[11] Despite this, some women with positive tests for APL antibodies have uncomplicated pregnancies without treatment.[11,12] Although other autoantibodies such as anticardiolipin or anti-β2-GP1 antibodies are associated with recurrent abortions, LA has been reported to be more specific for predicting clinical outcomes.[13] Most studies have been carried out in Caucasian populations; in this environment, literature is scarce on the association between APL antibodies and pregnancy loss. This study was to determine the prevalence of LA in women presenting with SA in Zaria, using Staclot® LA test for confirmation of LA.

Materials and Methods

It was a cross-sectional study carried out on a cohort of 100 consecutive patients with SA (defined as the termination of pregnancy before the gestational age of 28 weeks)[14] at the Gynaecology Clinic of ABUTH Shika-Zaria. An informed written consent was obtained from all participants and those who met the inclusion criteria were selected. An interviewer-administered questionnaire was used to collect sociodemographic, gynaecological and obstetric data from the respondents. Women were questioned regarding the number and timing of their abortions, family history of recurrent abortions, history of still births or intrauterine deaths, and history of thrombotic episodes. Women on anticoagulants, cervical incompetence, uterine abnormalities, diabetes mellitus, HIV, and other medical conditions were excluded from the study. Seven milliliter (mL) of venous blood was obtained from each participant following standard aseptic procedure. Exactly 4.5 mL of blood was dispensed into a bottle containing 0.5 mL of 3.2% of trisodium citrate with blood: anticoagulant ratio of 9:1 for coagulation studies. The remaining 2.5 mL of blood was dispensed into an EDTA bottle for determination of full blood count (FBC) and direct Coombs' test (DAT).

The FBC and DAT were done manually using the procedure described by Dacie.[15] Platelet poor plasma for coagulation tests was obtained by double centrifugation at 2500 g for 15 min.[15] Prothrombin time (PT), APTT, and Kaolin clotting time (KCT) were performed using STA– NEOPLASTINE®, PTT-LA (Diagnostica STAGO), and LUPAKCT®,[16,17] respectively. Samples with prolonged APTT were screened for the presence of an inhibitor by 50:50 mixture of patient and pooled normal plasma while Rosner index[15] was established for prolonged KCT. Failure of correction of prolonged APTT was confirmed using Staclot®-LA by diagnostic STAGO which is a phospholipid neutralization test. LA is confirmed when there was significant shortening of the APTT (difference of ≥8 s) or the Rosner index was >1.5. After 12 weeks, repeat LA tests for the four confirmed cases were positive; this ruled out transient LA.

Data were analyzed using Statistical Package for Social Sciences (SPSS) version 17.0 software (IBM). Quantitative variables were summarized using measure of central tendency, dispersion, and Student's *t*-test with *P* value set at 0.05. Chi-square test was used to demonstrate statistical significance.

Results

The mean age of the participants (±2 standard deviation) was 31.0 ± 3.8 years with a modal parity of 3 and a modal gestational age of 12 weeks. The past history of SA and confirmation of uncorrected prolonged APTT were shown in Table 1.

LA-positive patients had a higher median age and a higher gestational age pregnancy than the LA-negative patients. Patients with LA, compared with those without LA, had significantly prolonged PT, APTT, and KCT ($P \leq 0.001$, <0.001, and 0.01, respectively).

Table 1: Characteristics of the study participants (*n*=100)

Characteristics	Value
Mean age±2SD	31.0±3.8
Modal parity (range)	3 (0-12)
Modal gestational age in weeks (range)	12 (8-28)
History of previous SA	
0	32
1	28
2	19
>2	21
Uncorrected prolonged APTT	8
LA confirmed with Staclot®	4
LA confirmed with KCT	3

SD=Standard deviation; SA=Spontaneous abortion; APTT=Activated partial thromboplastin time; LA=Lupus anticoagulant; KCT=Kaolin clotting time

Table 2: Characteristics of lupus anticoagulant-positive and lupus anticoagulant-negative participants

Variables	LA positive (*n*=4)	LA negative (*n*=96)	*t*-test	*P*
Median age (years)	37.0	29.2	-	-
Median gestational age (weeks)	15.0	12.8	-	-
Mean PCV	32.5	31.6	0.435	0.67
Mean WBC	5.4	7.6	−1.344	0.18
Mean platelet count	240.7	258.8	−0.364	0.716
Mean PT	17.5	13.6	4.304	<0.001
Mean APTT	59.7	35.0	11.590	<0.001
Mean KCT	143.9	77.3	11.459	<0.001

KCT=Kaolin clotting time; APTT=Activated partial thromboplastin time; PT=Prothrombin time; WBC=White blood cells; PCV=Packed cell volume; LA=Lupus anticoagulant

Table 3: Comparison of past obstetric history between lupus anticoagulant-positive and lupus anticoagulant-negative participants

Past obstetric history	LA positive (*n*=4), *n* (%)	LA negative (*n*=96), *n* (%)	POR	95% CI
Preeclampsia/eclampsia	1 (25.0)	14 (14.6)	1.9	0.2-20.1
Normal	3 (75.0)	82 (85.4)		
Small for gestational age	1 (25.0)	9 (9.4)	3.2	0.3-34.3
Normal	3 (75.0)	87 (90.6)		
Had previous SA	3 (75.0)	66 (68.7)	1.4	0.1-13.6
No previous SA	1 (25.0)	30 (31.3)		

LA=Lupus anticoagulant; POR=Prevalence odds ratio; CI=Confidence interval; SA=Spontaneous abortion

Table 4: Comparison of the detection rate of Kaolin clotting time and Staclot®

KCT (*n*=100)	Staclot® (*n*=100)		Total
	Positive	Negative	
Positive	3	0	3
Negative	1	96	97
Total	4	96	100

McNemar test=1.000. KCT=Kaolin clotting time

However, there were no significant differences in their packed cell volume (PCV), white blood cell count (WBC), and platelet counts (P = 0.502, 0.271, and 0.121, respectively) [Table 2].

The prevalence odds ratio and 95% confidence interval for preeclampsia/eclampsia, small for gestational age deliveries, and previous SA for LA-positive and LA-negative groups were shown in Table 3.

There is no difference in the detection rate of STACLOT and KCT as P = 1.000 (McNemar Chi–square test) [Table 4].

Discussion

This study found a prevalence of LA to be 3% using KCT and 4% using Staclot® in women with SA in Zaria, northern Nigeria. This is comparable to the finding of Awodu *et al*.[18] working in southern Nigeria who reported a prevalence rate of 4.35% in women with recurrent SA s using KCT. However, this is lower than the prevalence of 13.5% reported by Al Samarrai in Iraq.[19] The differences might be due to genetic variations in the two populations or due to sampling methods used. Furthermore, the low prevalence of LA in this study could be explained by the fact that the diagnosis of LA in pregnancy is often very difficult due to changes in coagulation proteins.[20]

In this study, we found that the median gestational age for SA was 15 weeks. This is higher than for women without LA (12.8 weeks). This is different from the finding of Rai who reported that majority of SA in women with APL syndrome occur between 7th and 12th weeks of gestation.[21] In another separate study, it was observed that women with APL antibodies have an unusually high proportion of pregnancy losses within the fetal period (10 or more weeks of gestation).[22,23] The reason for this could be because, women with LA, in this study, were older than women without LA (37 years vs. 29.2 years). It has been shown that prevalence of LA, like other autoantibodies, increases with age.[24]

We also found that a woman with LA, although not statistically significant, was more likely to have previous bad obstetric histories such as preeclampsia/eclampsia, small for gestational age delivery, and previous SA than a woman without LA. These and other adverse pregnancy outcomes have been attributed to the presence of APL antibodies.[13]

The detection rate of APTT/Staclot® was not significantly different from that of KCT, 4% versus 3% (P = 1.000).

This is similar to the study by Kotila and Fasola[25] who found a detection rate of 4.8% and 3.2% for APTT and KCT, respectively. However, Olayemi and Halim[24] in Nigeria and Al Samarrai *et al*.[19] in Iraq reported KCT to be most sensitive in screening for LA.

One of the four cases of LA had positive DAT although the mean platelet counts, PCV, and WBC were not different from those without LA (P = 0.716, 0.850, and 0.182, respectively). This is at variance with Al Samarrai[19] who found reduction in platelet counts in patients with APA. Triplett[20] reported varying degrees of immune-mediated thrombocytopenia, and to some extent, autoimmune hemolytic anemia in patients with APL antibody positivity.

Our study is not without limitations. First, the cross-sectional study design coupled with the small sample size could not establish causality and also makes generalization of findings difficult. Second, even though LA has been recognized to be the most specific predictor of SA, not investigating for other APS antibodies limits the scope of detection of LA. Third, other adverse pregnancy outcomes and pregnancies beyond 28 weeks of gestation were not explored in our study.

Conclusion

Patients with LA were more likely to have a previous history of SA, preeclampsia/eclampsia, and small for gestational age delivery. There is no significant difference between the detection rate of APPT/Staclot® and KCT. Therefore, every woman presenting with adverse pregnancy outcomes and having uncorrected APTT following 50:50 mixing study with pooled control plasma should be routinely investigated for LA.

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Conflicts of interest

There are no conflicts of interest.

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