

Prevalence of IgG Anticardiolipin Antibody in Recurrent Pregnancy Loss-Sari (2000-2003)

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Abstract: Recurrent spontaneous abortion and fetal loss are common in obstetrics and gynecology (60% of pregnancy loss). Immunological factor suspected to have a major role in These problem. Recent studies suggest association of antiphospholipid antibodies, especially anticardiolipin antibodies (aCL) with recurrent pregnancy loss. Different studies report different association (5-51%), we determine the prevalence of aCL in recurrent, pregnancy loss in Sari during 2000-2003. In a descriptive study women with a history of at least two pregnancy loss without any known etiology like hormonal imbalance, systemic hypertension, diabetes, infection, hyperthyroidism and, anatomical abnormality in genital tract were studied. 512 cases, for presence of aCL in serum were investigated. The patient sera were tested by Enzyme Linked Immunosorbent Assay (ELISA) method. Commercial standard ELISA kit from GENESIS company were used, According the kit procedure, 5 to 11 U mL⁻¹, 11 to 13 U mL⁻¹ and >13 U mL⁻¹ results of measurement were, respectively consider as the negative (normal), borderline and positive values. The prevalence of aCL determined and Confidence interval (CI) were estimated. 512 women with age range of 18-40 (28.02±5.66) were studied. aCL was detected in sera of 57 (11.1%) cases, with confidence interval of 2.7 (11.1±2.7) 447 (87.4%) cases were negative for aCL and in 8 (1.5%) were intermediate. 49.1 of pregnancy loss with positive aCL were in first trimester of pregnancy and 50.9 in second and third trimester with odd's ratio of 3.1. The prevalence of aCL in recurrent abortion and fetal loss in Sari are not high and it is higher in second and third trimester.

Key words: Anticardiolipin antibody, recurrent abortion recurrent fetal loss

INTRODUCTION

Unwanted recurrent pregnancy Loss is one of the known problem in medicine. Depend to the time of event; this phenomenon refers to the two terms, abortion (end of pregnancy, from beginning of gestation to 20 week) and fetus loss (end of pregnancy, from 20 weeks). The incident of this problem consists of 30% of all pregnancy (Williams, 1997). The causes of abortions and fetus deaths are different and many factors like hormonal, genetically, uterus anatomical disorders, systemic hypertension, Diabetes, hyperthyroidism and infectious diseases have been well known to cause this problem (Heliodelima *et al.*, 1993). In spite of these factors, the causes of many (60%) spontaneous abortions remain unknown. Apparently, immunological factors are involved in these cases (Sulani *et al.*, 2003) (80%). The elevation of anti-phospholipid antibody Plasma Level (aPL) has been reported to be one of these factors (Chakrabarti *et al.*, 1999; Kutteh, 1997). These antibodies are different group of auto-antibody that formed against the phospholipoproteins with negative charges in body.

These antibodies are included anti-Cardiolipin (aCL), Lupus Anti-coagulant (LA), anti-beta-2-glycoprotein (aβ-2-GP-1), anti-Phosphoserin (aPS), anti-Phosphatidyl Glycerol (aPG), anti-Phosphatidic Acid (aPA) anti-phosphatidyl inositol (Chakrabarti *et al.*, 1999; Kutteh, 1997; Branch, 1998). Among these antibodies, anti-cardiolipin and Lupus anti-coagulant are known to have more critical and significant role to form the spontaneous abortions and fetal losses (Branch, 1998; Vinatier *et al.*, 2001). For the women with recurrent abortions with no reason and the positive aCL test, aspirin with low dosage, prednisolon, heparin and immunoglobulin are given as the drugs of choice (Williams, 1997).

The intensity of aCL effect on spontaneous recurrent abortions has been differently reported by several studies. In one of them, 42.8% of pregnant women have positive serum level of anti-cardiolipin. These patients had at least one miscarriage in 2nd or 3rd trimesters (Kiwamn *et al.*, 1991).

In other study, 50% of pregnant women with recurrent abortion showed the positive level of this antibody (Heliodelima and Fernands, 1993;

Howard *et al.*, 1987; Blumenfeld *et al.*, 1991). A more comprehensive study reported the measurement of the positive aCL plasma level in 15.1%, pregnant women with abortion in the 1st trimester and 41.3% in the 2nd and 3rd trimesters (Masahike, 1998).

Certain sources reported the positive level of this antibody for 2-5% of pregnancy with a normal child birth and 7-17% of pregnancy with fetus loss (Branch, 1998; Vinatier *et al.*, 2001).

In Iran, the only report has been presented was in 6th congress of immunology (Soutoudeh *et al.*, 2002. This study showed the significant difference between aCL and antinuclear antibody level in patients in comparison with control group. This research evaluated the selected pregnant women with at least two losses of pregnancy in the years 2000 to 2003 which they were admitted by the different clinics in Sari to determine the positive cases of Anti-cardiolipin antibody.

MATERIALS AND METHODS

In a descriptive study prevalence rate of aCL were investigated on patients with recurrent abortion or fetal loss in sari gynecological clinics. Regarding prevalence rate of aCL (5-51%) in similar study in the world and with $d = 0.05$ and $Z = 1096$, sample size were determined.

Selected patients had at least two loss of pregnancy without considering the time of fetus death during Their pregnancy. They worked out by gynecologist for known etiology of recurrent pregnancy loss. The patients without known etiology such as hormonal and uterus abnormality, systemic hypertension, hyperthyroidism, diabet, acalampsia, pre-ectampsia, touch syndrome were included in This study. Blood samples were taken at the most 3 month since their last abortion. serum of each sample was separated and it was kept at -20°C . A commercial standard ELISA kit produced by Genesis company (UK) was used to determine the anti-cardiolipin antibody-IgG type. According the kit procedure, 5 to 11 U mL^{-1} , 11 to 13 U mL^{-1} and $>13 \text{ U mL}^{-1}$ results of measurement were, respectively consider as the negative (normal), borderline and positive values.

RESULTS

The patients had the age average of 28.02 ± 5.66 with the range of 18 to 40. 245 numbers of them (47.8%) had no child and the rest (52.2%) had at least one child. The later group had experience of two or more recurrent abortion. The 370 cases (72.3%) had abortion in 1st trimester and 142 patients had this in 2nd or 3rd trimesters during their pregnancy. In total, 57 cases (11.1%) showed the positive

Table 1: The patient's distribution with recurrent abortion, according the different stages of pregnancy, years 2001-2004

aCL/pregnancy stages	Negative	Borderline	Positive
1st trimester	337 (75.4%)	5 (62.5%)	28 (49.1%)
2nd, 3rd trimesters	110 (24.6%)	3 (37.5%)	29 (50.9%)
Total	448	8	57

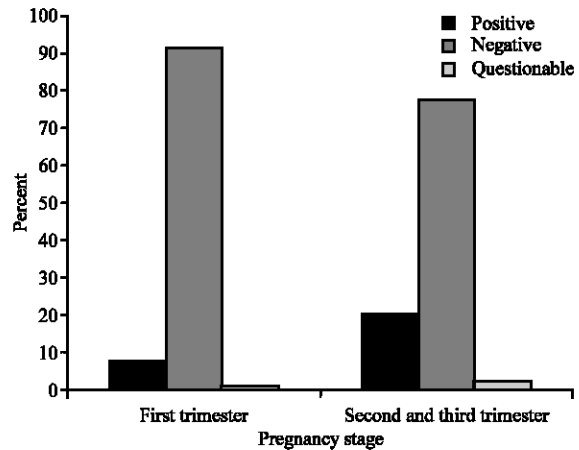


Fig. 1: Distribution of 512 patients with recurrent abortion and fetal loss according to the IgG aCL level in plasma

with confidence interval value of 2.7 (2.7 ± 11.1), 8 cases (1.5%) were suspect and 447 cases (87.4%) showed the negative anti-cardiolipin antibody values. 49.1% of the positive cases showed to have abortion in 1st trimester and 50.9% had recurrent abortion and fetus loss in 2nd and 3rd trimesters during their pregnancy (Table 1).

With respect of the calculated odd's ratio for the pregnancy process, this study showed that the risk of fetus death and abortion in 2nd and 3rd trimesters is 3.1 greater than the 1st trimester. Figure 1 shows that 7.5% of total abortion and fetus death occurs in the 1st trimester and 20.4% appear in the 2nd and 3rd trimesters.

DISCUSSION

Anti-cardiolipin antibody was found in serum of 11.1% of the patients with abortion in Sari.

49.1 percent of the positive cases had fetus death during their 1st trimester and 50.9% of them had this disorder in their 2nd and 3rd trimesters.

The existence of anti-phospholipid antibody as the cause of recurrent abortion has been reported by several epidemiologic studies with frequency of 5 to 51% (Vinatier *et al.*, 2001). Table 2 shows the plasma level percentages of this antibody presented by these studies.

In a study (Aoki) which was carried out on the 334 women with the record of fetus death and without other type of autoimmune diseases (Aoki *et al.*, 1993).

Table 2: The percentages of anti-phospholipid antibody plasma level in patients with recurrent abortions and fetus death in the different studies

Source of research	aCL (%)	No. of patients
Petri (1987)	11	44
Howard (1987)	48	9
Barbai (1988)	8	49
Parazzini (1991)	19	220
Parke (1991)	7	80
Out (1991)	5	102
Blumenfeld (1991)	50.7	67
Rai (1995)	5.5	500
Yetman (1996)	17.3	866
Branch (1997)	16	147

Fourteen percent patients showed that had at least one type of antibodies which are formed against the six main phospholipid compounds in body. Anti-cardiolipin (aCL), anti-phosphatidic acid (apA) and antiphosphatidyl glycerol (apG) have been found to be the most common cases among other antibodies (Aoki *et al.*, 1993). In a study, Yetman and Kutteh measured 5 anti-phospholipid antibodies of 866 women with recurrent abortion, fetus death and 388 women with normal pregnancy. The positive cases of anti-cardiolipin were 17% of cases and 4% of control group (Yetman and Kutteh, 1996). The same study was carried out by Ogasawa and the measurement showed that 18% of patients and 9% of normal women were respectively positive (Branch *et al.*, 1997). In Iran, the only study on anti-phospholipid antibodies was presented in 6th Iranian congress of immunology (Soutoudeh *et al.*, 2002). This study showed that anti-cardiolipin and anti-nuclear antibody was significant in cases with recurrent abortion and fetus death in comparison with the control group (Soutoudeh *et al.*, 2002). Meanwhile, some study even showed that the positive cases are between 48 to 50% (Table 2) (Howard *et al.*, 1987; Blumenfeld *et al.*, 1991). The time of anti cardiolipin effect to end a pregnancy have been differently reported by the different researches but some studies believe that this effect appeared more in the 1st trimester (Branch *et al.*, 1992; Oshiro *et al.*, 1996) and some in the 2nd and 3rd trimesters (Barbui *et al.*, 1988; Parke *et al.*, 1991; Parazzini *et al.*, 1991; Petri *et al.*, 1987).

This study showed that this effect appeared in the 2nd and 3rd trimesters with the relative high risk value of 3.1.

In spite of, the relationship between anti-phospholipid antibodies and abortion has been proved by many scientists and studies but all showed a variation (5 to 51%) which it seems due to the many reasons as given below:

- Definition of recurrent abortion and fetus loss is different in literatures. Some studies refer the diagnosis recurrent abortion after the 2nd abortion including this research, but the epidemiological literatures refer to the 3rd abortion.

- The assay for determination of anti-phospholipid antibodies is not standardized. For instance, in a study IgM and IgG type were measured against six epitope of phospholipids with 59% of the positive cases (Matzner *et al.*, 1994). In other study, only IgG level was measured and the positive cases were 16% (Branch *et al.*, 1997).

In this study, IgG type was also measured and the positive cases were 11.1%.

- Exclusion and inclusion criteria of patients are different in different studies. For instance, in some, any autoimmune diseases is excluded from research and in other, the only patients without any known cause of abortion were evaluated.

Therefore, the obtained prevalence of aCL (11.1%) by this research was similar to other study done under the same condition.

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