

## CLINICAL CONSIDERATIONS IN RECURRENT MISCARRIAGE

M. Biriş<sup>1</sup>,  
A. Raţiu<sup>1</sup>,  
D.C. Crişan<sup>1</sup>,  
D. Păscuţ<sup>1</sup>,  
A. Motoc<sup>1</sup>

### SUMMARY:

*The aim of this study was to examine the benefit of low molecular weight heparin and low dose aspirin in treatment of recurrent miscarriage.*

*Recurrent miscarriage is a heterogeneous condition defined as three consecutive miscarriages before 10 weeks of amenorrhea and affects 1%-2% of women of reproductive age. The suggested causes include prothrombotic states, structural uterine anomalies, chromosomal anomalies, endometrial and endocrinological defects. In 50% of the cases neither of the above can be identified.*

*We performed a prospective study on 26 consecutive patients between January 2002-July 2009, looking for preconception counseling or prenatal care at „Dr. Dumitru Popescu” Clinic Hospital, Timișoara. All women had history of recurrent pregnancy loss, defined as at least three consecutive miscarriages. In the same period in our hospital took place 24906 deliveries.*

*Treatment with low molecular weight heparin in prophylactic doses and low dose aspirin improved live births rate even when proper diagnosis of thrombophilia cannot be performed.*

**Key Words:** recurrent miscarriage, thrombophilia, low molecular weight heparin.

### CONSIDERAȚII CLINICE ÎN AVORTUL RECURENT

#### Rezumat:

*Scopul acestei lucrări a fost de a examina beneficiile tratamentului cu heparină cu greutate moleculară mică și aspirină în doză mică în tratamentul avortului recurent.*

*Avortul recurent este o condiție heterogenă, definită ca cel puțin trei avorturi spontane consecutive înainte de 10 săptămâni de amenoree și afectează 1-2% din femeile de vârstă reproductivă. Cauzele posibile includ statusul protrombotic, anomaliile structurale uterine, anomaliile cromozomiale și deficitel endometriale sau endocrinologice. În aproximativ 50% din cazuri, nici una dintre acestea nu poate fi identificată.*

*Am efectuat un studiu prospectiv pe 26 de paciente consecutive în perioada ianuarie 2002 - iulie 2009, dispensarizate pentru consiliere preconceptuală sau prenatală la Spitalul Clinic “Dr. Dumitru Popescu” Timișoara. Toate pacientele au avut istoric de avort recurent, definit ca trei avorturi consecutive până la 10 săptămâni de amenoree. În aceeași perioadă, în spitalul nostru au avut loc 24906 nașteri.*

*Tratamentul cu heparină cu greutate moleculară mică în doză profilactică și aspirină în doză mică a îmbunătățit rata de nou născuți vii, chiar și când un diagnostic adecvat de trombofilie nu a putut fi stabilit.*

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1. - 2nd Department of Obstetrics-Gynecology, “Victor Babeș” University of Medicine and Pharmacy, Timișoara

## INTRODUCTION

Miscarriage represents an important issue for our clinic. Approximately 15% of all pregnancies will be unsuccessfully. Recurrent miscarriage (RM) is a condition defined as three consecutive miscarriages before 10 weeks of amenorrhea and affects 1%-2% of women of reproductive age [1]. Up to 5% have 2

recurrent losses. These sporadic miscarriages are the commonest complication of pregnancy and are mainly due to chromosomal abnormalities in the fetus.

RM is a heterogeneous condition, with several etiological factors such as prothrombotic states, structural uterine anomalies, chromosomal anomalies, and endocrinological defects. In up to 80%, however, the

**Correspondence to:** Marius Biriş, e-mail: marius308@yahoo.com

underlying cause is not apparent and the condition is therefore considered unexplained [1].

The haemostatic system has a major importance in the success of pregnancy and the process of implantation, and placentation. Women with thrombophilia are at increased risk for thrombosis during pregnancy and adverse maternal and fetal sequelae [2]. Implantation of the fertilized egg into the uterine decidua establishes a contact between the fetus, the placenta and the maternal circulation. This contact between placenta and maternal circulation is crucial for the success of pregnancy. Prothrombotic changes and thrombosis may interfere with these processes leading to miscarriage. This may explain many cases of previously unexplained RM.

Preeclampsia, abruptio placenta, intrauterine growth restriction (IUGR) and intrauterine fetal death (IUFD) greatly contribute to maternal and fetal morbidity and mortality. All of them may be associated with abnormal placental vascularisation and perturbations of homeostasis leading to deficient maternal-fetal circulation [3, 4].

Thrombophilias are inherited or acquired conditions which predispose an individual to thromboembolism.

Deficiencies of protein S, C and antithrombin (AT III) are rare and each of them is found in about 3% of patients with thrombosis. Recently, three important inherited thrombophilias were discovered which are responsible of the majority of thromboembolic events in patients with otherwise no apparent risk for thrombosis. Resistance to activated protein C caused by an adenine 506 guanine (A506G) mutation in factor V (factor V Leiden) has been linked with an increased risk for venous thromboembolism [5-7]. Heterozygosity for the factor V Leiden mutation is found in about 5% of the population and the mutation is responsible of 20–30% of venous thromboembolism events. A recently described guanine 20210 adenine mutation in prothrombin is associated with higher plasma prothrombin concentrations and increased risk for venous thromboembolism [5] and cerebral-vein thrombosis [9]. Homozygosity for the cytosine 677 thymine (C677T) mutation in methylenetetrahydrofolate reductase (MTHFR) results in decreased synthesis of 5-methyltetrahydrofolate, the primary methyl donor in the conversion of homocysteine to methionine and the resulting increase in plasma homocysteine concentrations is a risk factor for thrombosis [10, 11]. The mutation is responsible for reduced MTHFR activity and is the most frequent cause of mild hyperhomocysteinemia and can be found in 5–15% of the population [1].

Homocysteine is an independent risk factor for atherosclerosis, stroke, peripheral vascular disease and cardiovascular diseases [12, 13]. Homocysteine concentrations are affected by nutrition. A deficiency in folate, B-6, and/or B-12 causes elevation of homocysteine. Homocysteine concentrations are also affected by genetics such as cystathionine beta-synthase deficiency [14] and C677T MTHFR gene mutation [11]. Hyperhomocysteinemia promotes vascular damage by several mechanisms. Many of the endothelial vascular changes associated with hyperhomocysteinemia can be found in preeclampsia [15-19].

The antiphospholipid syndrome (APS), is defined as the presence of lupus anticoagulant and/or anticardiolipin antibodies with recurrent miscarriage, thrombosis, preeclampsia, IUGR and placental abruption. The most specific clinical characteristics are thrombosis, recurrent miscarriage and fetal loss in the second and third trimester and autoimmune thrombocytopenia [20-22].

The aim of our study was to examine the benefit of low molecular weight heparin (LMWH) and low dose aspirin in treatment of RM even when thrombophilias couldn't be proved.

## MATERIAL AND METHODS

We performed a prospective study on 26 consecutive patients between January 2002-July 2009, looking for preconception counseling or prenatal care at „Dr. Dumitru Popescu” Clinic Hospital, Timișoara. All women had history of recurrent pregnancy loss, defined as at least three consecutive miscarriages. In the same period in our hospital took place 24906 deliveries.

The study was approved by our local hospital ethics committee. All patients were fully informed of the aim of the study and of the proposed treatment regimen, before definitive study enrollment; informed consent was obtained from all participants.

We divided these patients into two groups. Group A formed by 11 cases with positive preconception thrombophilia testing: AT III and protein C deficiency (chromogenic test), protein S deficiency (coagulation test), factor V Leiden, prothrombin and MTHFR gene mutation (real-time PCR), APS lupus anticoagulant (coagulation test) and anticardiolipin antibodies (ELISA). Group B represented by 15 pregnant women with first trimester pregnancy between 5-8 weeks without thrombophilia testing. Both groups had genitor karyotyping, uterine abnormalities, endometrial and endocrinological defects excluded.

**Table 1.** Demographic characteristics of groups

	GROUP A	GROUP B
<b>Number</b>	11	15
<b>Age</b>	27.36±4.27	28.33±3.63
<b>BMI</b>	26±3.68	25.26±2.89

**Table 2.** Type of thrombophilia in group A

Protein S deficiency	0
Protein C deficiency	0
AT III	0
Factor V Leiden mutation	0
Protrombin gene mutation	0
MTHFR	4
Antiphospholipid syndrome	7
<b>Total</b>	<b>11</b>

All women received aspirin, 75mg daily when intrauterine pregnancy was confirmed by transvaginal ultrasonography and low molecular weight heparin in prophylactic doses after documenting the viability of the embryo. The first three cases from study group (two from group A and one from group B) received Dalteparin and the rest Enoxaparin. The aspirin was administered until 24 weeks and LMWH until term and 6 weeks postpartum in thrombophilia group.

We assessed the occurrence of early or late miscarriage, intrauterine growth retardation, unexplained intrauterine fetal death, preeclampsia, abruptio placentae, venous thromboembolism, the possible complications of therapy (severe antenatal bleeding,

postpartum hemorrhage, thrombosis, and thrombocytopenia) and the pregnancy outcome. We compared them between the two groups using student t test, statistical significance was considered at a P value less than 0.05.

## RESULTS

The demographical characteristics between the two groups were similar (Table 1).

Group A was tested for thrombophilia and we found 7 cases of antiphospholipid syndrome and 4 cases of homozygote genotype of metilentetrahydrofolat reductase deficiency (MTHFR), two with mutation C677T and two with adenine 1298 cytosine (A1298C) mutation (Table 2).

Group B from our study was not tested for the presence of thrombophilia because the patients referred to our clinic when already pregnant. All women from both groups received also Folic Acid 5mg daily.

The use of low-molecular-weight heparin Enoxaparin combined with aspirin was associated with an impressively higher rate of healthy live births in all the women but also in each of the two groups, 9 in group A with 1 preterm birth and 11 in group B with two preterm births. We found no statistical significant difference between the two groups regarding to outcome of the pregnancy. We used Dalteparin in three cases but 2 ended with early abortion and one with a late miscarriage.

In our study groups we have not found a significant difference between the cases complicated by intrauterine growth retardation and there were not any case of intrauterine fetal death or abruptio placentae (Table 3).

**Table 3.** Complications and outcome of pregnancies

Complications and outcome of pregnancies	Group A	Group B	p value
Early abortion	2	3	0.3
Late abortion	0	1	0.5
IUGR	1	1	0.5
Intrauterine fetal death	0	0	-
Preeclampsia	1	0	0.3
Abruptio placentae	0	0	-
Venous thromboembolism	0	0	-
Preterm live birth	1	2	0.5
Term live birth	8	9	0.3

**Table 4.** Complications of therapy

Complications of therapy	Group A	Group B
Severe antenatal bleeding	1	0
Postpartum hemorrhage	0	1
Thrombosis	0	0
Low platelet count	0	0
Injection site bruising	3	4
<b>Total</b>	<b>4</b>	<b>5</b>

We have not observed any case of heparin-induced thrombocytopenia, abnormal skin reactions, or clinical manifestation of spontaneous bone pain among the women treated with LMWH. No case was seen of digestive intolerance to low-dose aspirin either. There were one case of hemorrhage linked to an incomplete abortion and one postpartum hemorrhage in group B; slight bruising at the injection sites was noted in both groups (Table 4).

## DISCUSSIONS

Prospective studies have demonstrated an increased prevalence of antiphospholipid antibodies (aPL) among women with first trimester RM [2]. Some 15% of women with RM have persistently positive tests for aPL.

Brenner et al. [23] tested women with 3 or more first trimester losses, 2 or more second trimester losses or one or more third trimester loss. The FV Leiden mutation was more frequent in the fetal loss group compared to controls. Studies regarding the association of MTHFR and recurrent pregnancy loss are contradicting with some who negate an association between MTHFR and recurrent abortions [23, 24], and others who find such association [25, 26].

Rai et al. [27] performed a randomized controlled trial of aspirin and aspirin plus heparin in pregnant women with RM associated with antiphospholipid antibodies. The rate of live birth in patients treated with aspirin and heparin was 71% compared to 42% with aspirin alone ( $p < 0.01$ ). Still, 25% of successful pregnancies were delivered prematurely. Treatment consisted of

prophylactic heparin (5,000 IU b.i.d.), or LMWH, and low-dose aspirin (0.1 g per day). The data for inherited thrombophilias is even more limited and no controlled trials exist.

In our study, the use of association of low molecular weight heparin in prophylactic doses and low dose aspirin improved significantly pregnancy outcome in both groups and appeared to be safe.

Most remarkable, patients with thrombophilia markers had live births at a similar frequency as patients without those parameters [28]. No severe side effects of LMWH were seen. While treatment with heparin and aspirin from early pregnancy has been shown to significantly increase the live birth rate, the incidence of late pregnancy complications still remains possible [29].

## CONCLUSIONS

Treatment with low molecular weight heparin in prophylactic doses and low dose aspirin reduced significantly pregnancy complications and improved live births rate in both groups, with thrombophilia testing and without thrombophilia testing.

In pregnant women with history of recurrent pregnancy loss, when proper diagnosis of thrombophilia cannot be performed, it seems to be safe and useful to administer low molecular weight heparin in association with low dose aspirin in order to maintain a pregnancy and improve neonatal outcome. The potential mechanism of action of LMWH in early and late abortions warrants further study.

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