# Thrombophilia and Recurrent Pregnancy Loss
## Is heparin still the drug of choice?

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**ABSTRACT:** The association between thrombophilia and recurrent pregnancy loss (RPL) has become an undisputed fact. Thorombophilia creates a hypercoaguable state which leads to arterial and/or venous thrombosis at the site of implantation or in the placental blood vessels. Anticoagulants are an effective treatment against RPL in women with acquired thrombophilia due to antiphospholipid syndrome. The results of the use of anticoagulants for treating RPL in women with inherited thrombophilia (IT) are encouraging, but recently four major multicentre studies have shown that fetal outcomes (determined by live birth rates) may not be as favourable as previously suggested. Although the reported side-effects for anticoagulants are rare and usually reversible, the current recommendation is not to use anticoagulants in women with RPL and IT, or for those with unexplained losses. This review examines the strength of the association between thrombophilia and RPL and whether the use of anticoagulants can improve fetal outcomes.

*Keywords:* Thrombophilia; Recurrent Abortions; Spontaneous Abortions; Heparin, Low-Molecular-Weight; Aspirin.

**الملخص:** إن العلاقة بين أهبة التخثر وفقدان الحمل المتكرر هي حقيقة واقعة. تؤّدي أهبة التخثر إلى زيادة التخثر في الدم و بالتالي إلى خثار في الشرايين أو الأوردة في موقع العلوق أو في الأوعية الدموية للمشيمة. تعمل الأدوية المضادة للتخثر بفعالية في علاج فقدان الحمل المتكرر عند السيدات اللواتي يعانين من أهبة التخثر المكتسبة الناتجة عن ظاهرة الأجسام المضادة الشحمية الفوسفورية. إن نتائج الدراسات التي تم فيها استعمال الأدوية المضادة للتخثر لمعالجة فقدان الحمل المتكرر كانت مشجعة للغاية حتى صدور أربعة دراسات كبرى متعددة المراكز بينت أن نسبة المواليد الأحياء ربما لا تكون مشجعة كما كان يعتقد سابقا. على الرغم من أن المضاعفات والأعراض الجانبية لتلك الأدوية نادرة الحدوث وليست خطيرة فان تلك الدراسات لا توصي باستعمال الأدوية المضادة للتخثر لعلاج الأمهات اللواتي يعانين من فقدان الحمل المتكرر أهبة التخثر الوراثية لعلاج فقدان الحمل المتكرر الذي لا يعرف له سبب.

**مفتاح الكلمات:** أهبة التخثر؛ الحمل المتكرر؛ الهيبارين ذو الوزن الجزيئي المنخفض؛ الأسبرين المعتاد.

RECURRENT PREGNANCY LOSS (RPL), either early or late in the gestational period, is a serious problem and has both psychological and social impacts on the women who suffer from it. In some cases, it may lead to divorce or other social problems.

Miscarriage is common, with most studies showing that the incidence of this complication occurring before 20 weeks' gestation varies between 8–20%, with 80% of these occurring in the first 12 weeks of pregnancy.[1] The real rate of miscarriage may be much higher than reported, since many women miscarry before realising that they are pregnant. In one study, human chorionic gonadotrophin was checked daily from the expected time of ovulation until the next menstrual period in order to detect a pregnancy as early as possible; this yielded a miscarriage rate of 31%.[2]

Habitual or recurrent miscarriage (RM) is defined as the loss of three or more consecutive and clinically-recognised pregnancies before 20 weeks' gestation; this affects 1–2% of women.[3] This incidence increases to 5% when it is defined as a loss of two or more clinically-recognised pregnancies before 20 weeks' gestation.[4] RPL may be classified as early (losses at or before 20 weeks' gestation) or late (losses after 20 weeks' gestation). Patients may be classified as suffering from primary RPL when they have never had a live birth or from secondary RPL when they have had recurrent losses following

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a successful pregnancy. RPL has now been deemed a major cause of female infertility.[5]

Thrombophilia is a common cause of RPL and may be seen in 40–50% of cases.[6,7] Pregnancy is a hypercoaguable state and if the pregnancy is affected by thrombophilia, the hypercoaguable state becomes worse and may impair blood flow through the maternal veins, leading to deep vein thrombosis, and clots in the placental blood vessels, leading to fetal growth restriction and/or fetal demise.[8,9] Due to this fact, anticoagulants have become very popular for treating RPL.

The aim of this review is to find the strength of the association between thrombophilia and RPL and whether the use of anticoagulants can improve fetal outcomes.

## Thrombophilia

Thrombophilia is a term which describes the increased tendency of excessive blood clotting. It is a normal phenomenon during pregnancy, where there is an increase in most clotting factors, such as factor VIII, Von Willebrand factor, platelets, fibrinogen and factor VII. During pregnancy, there is also an increase in prothrombin fragment 1 + 2 and D-dimer.[10,11]

When investigating patients with RPL, it is very important to exclude other possible causes of the losses, such as uterine malformation; diabetes mellitus; connective tissue diseases such as systemic *lupus erythematosus* (SLE); chromosomal abnormalities, and thyroid dysfunction.[12–15]

Currently, many clinicians treat RPL—either associated with all types of thrombophilia or unexplained—with low-molecular-weight heparin (LMWH) combined with low-dose aspirin (LDA). This treatment became popular in the late 1990s, after Sanson *et al.* reported that thrombophilia is associated with the high risk of fetal loss in early and late pregnancy.[16] Thrombophilia is either inherited, acquired or a combination of both.

### INHERITED OR GENETIC THROMBOPHILIA

In inherited or genetic thrombophilia, there is usually a family history of excessive clotting. More commonly, the diagnosis is based on the demonstration of a gene mutation such as a Factor V Leiden (*FVL*) mutation (C677T), a hyperhomocysteinaemia mutation (A506G), a prothrombin mutation (G20210A) or prothrombin II (PTII) mutation, or a protein S and/or C deficiency.

Clinical studies suggest that hypercoagulation is the main underlying pathophysiological mechanism which leads to uteroplacental insufficiency and, subsequently, pregnancy loss. It is believed that inherited thrombophilia (IT) impairs the placental function by causing arterial and/or venous thrombosis at the maternal-fetal interface. It is also believed that activated protein S and protein C inhibit the action of certain clotting factors, such as factors V and VIII. This shows that proteins S and C act as anticoagulants. If the actions of these proteins are reduced, the inhibition of the clotting mechanism is removed and, subsequently, placental arterial and venous thrombosis may occur; this mechanism might be the basis of RPL associated with thrombophilia.

When there is a mutation of the *FVL* gene (arginine amino acid is substituted by glutamine amino acid at position number 506 of factor V), this may result in the formation of a protein which is resistant to the action of activated protein C, called anti-protein C (aPC). The aPC removes the inhibitory effect of protein C on the clotting mechanism and enhances the conversion of prothrombin to thrombin, subsequently enhancing the formation of clots.[17–19] This absent or reduced activity of antithrombin leads to increased levels of thrombin and clot formation.

A mutation of the prothrombin gene (*G20210A*) will facilitate the formation of thrombin and clot formation in heterozygous individuals, who have a two-fold higher risk of clotting in comparison to non-carriers. Women with hyperhomocysteinaemia show a folic acid deficiency, also resulting in a two-fold increase in clotting within homozygous women.[20–23]

The exact mechanism by which IT causes implantation failure and subsequent RPL is unclear. It has been suggested that thrombophilia may lead to a syncytiotrophoblast invasion of the maternal blood vessels, which in turn leads to the formation of microthrombosis at the site of implantation, resulting in implantation failure and RPL.[24]

In a study of the effect of ethnicity on RM and IT, Baumann *et al.* found that in a uniform ethnic group the prevalence of various congenital thrombophilic

markers did not differ. Thus, when investigating a multi-ethnic cohort of women, the prevalence of hereditary thrombophilia may differ due to the fact that the basic prevalence in different ethnic groups varies.[25] The prevalence of thrombophilia in the general population varies from 1.1% in Lebanon to 2.5% in India.[26,27]

In acquired thrombophilia, antiphospholipid syndrome (APS) can be due to either lupus anticoagulant antibodies or anticardiolipin antibodies, as seen in women with SLE. In APS, the body's immune system recognises the phospholipids, which are a part of the cell membrane, as a foreign substance and thus produces antibodies against them. However, other studies have shown that antiphospholipid antibodies (aPL) often act against a protein cofactor called β2-glycoprotein 1. This protein cofactor helps the aPL to adhere to the phospholipids in the cell membrane.[28] The aPL consist of 20 antibodies, but only the lupus anticoagulant and anticardiolipin antibodies (immunoglobulin G and immunoglobulin M, but not immunoglobulin A [IgA]) have been shown to be of clinical significance.[29,30] In one study, 55% of women with RPL tested positive for aPL.[31]

In women with SLE, adverse live-birth outcomes were significantly associated with positititve anticardiolipin IgA and anti-beta 2 glycoproteins.[32] The mechanism by which APS causes implantation failure and subsequent RPL is unclear. It has been suggested that, as in thrombophilia, APS may lead to a syncytiotrophoblast invasion of the maternal blood vessels, leading to the formation of microthrombosis at the site of implantation, resulting in implantation failure and RPL.[24]

Histopathological examinations of the placentas in women with APS showed thrombosis, acute atherosis, a decreased number of syncytiovascular membranes, an increased number of syncytial knots and obliterative arteriopathy. These placental changes, although common, are not specific to APS.[33]

It has been suggested that in women with APS and RPL, the presence of aPL during pregnancy is a major risk factor for adverse fetal outcomes.[34] In a recent study, the incidence of aPL was 27.8% in couples with RPL.[35]

#### COMBINED THROMBOPHILIA

Combined thrombophilia (which is either a combination of acquired and IT, or a combination of more than one inherited thrombophilic gene defect) has been identified by several researchers as a cause of both early and late RPL; however, the frequency of combined thrombophilia is not clear.[36,37]

## Low-Molecular-Weight Heparin

Heparin exerts an effect on women with thrombophilia through various mechanisms; it potentiates the antithrombin effects of the condition, thus preventing clot formation.[38]

Heparin also binds to the aPL, rendering them inactive; this is important as these antibodies adhere to the cell surface and impede the differentiation and invasiveness of the cytotrophoblasts. It has been reported that low-molecular-weight heparin (LMWH) reduces the binding of the aPL to the trophoblast cells, subsequently restoring the cytotrophoblasts' invasiveness and differentiation.[39] Unfractionated and LMWH reduce E-cadherin protein expression in rat pregnancies. This may enhance the trophoblast invasion in patients with pregnancy loss.[40]

The treatment of women with APS and RPL with heparin is beneficial because complement activation is essential for the aPL to induce fetal damage; heparin, whether fractionated or LMWH, inhibits the complement activation *in vivo* and *in vitro* in pregnant mice, thus preventing fetal damage.[41]

In the absence of aPL, LMWH induces a potentially detrimental proinflammatory and anti-angiogenic profile in the trophoblast. In the presence of aPL, single-agent LMWH may be the optimal therapy to counter the trophoblast inflammation, however it also induces an anti-angiogenic response.[42]

## Treatment of Women with Antiphospholipid Syndrome and Recurrent Pregnancy Loss

The evidence in the literature favours treating aPL-positive women with APS who are suffering from RPL. The Cochrane review by Empson *et al.* was conducted to assess the efficacy of all treatment

options in order to improve pregnancy outcomes in aPL-positive women suffering from RPL.[43] The studies reviewed were either randomised or quasi-randomised studies of pregnant women with a history of RPL and aPL. The selection criteria (women with RPL and APS with positive aPL) (N = 849) were fulfilled in 13 studies. They found that combining unfractionated heparin with LDA (two trials; n = 140) resulted in a significant reduction in pregnancy loss when compared to a trial in which patients used LDA alone (relative risk [RR] = 0.46; 95% confidence interval [CI]: 0.29–0.71). One trial (N = 98) compared pregnancy loss outcomes when LMWH was combined with LDA in one group and LDA was given alone in another group. This did not show a significant reduction in pregnancy loss rate (RR = 0.78: 95% CI: 0.39–.57). One trial (N = 50) failed to show any advantage in giving high-dose or low-dose unfractionated heparin. The use of LDA alone was reported in three trials (N = 135) and these showed no significant reduction in pregnancy loss (RR = 1.05; 95% CI: 0.66–1.68). Treatment with prednisone and LDA was reported in three trials (N = 286). The results showed a significant increase in prematurity with LDA alone in comparison to a combination of heparin with LDA, and both of these treatment options were compared to a placebo. In fact, there was an increase in gestational diabetes. Intravenous immunoglobulin and/ or unfractionated heparin and LDA were used in two trials (N = 58). This was associated with a higher frequency of pregnancy loss and premature deliveries when compared to unfractionated heparin or LMWH combined with LDA (RR = 2.51; 95% CI: 1.27–4.95). When compared to LDA alone

and prednisone alone, intravenous immunoglobulin (one trial; N = 82) showed no significant difference in outcome. The authors concluded that the combination of unfractionated heparin and LDA may reduce pregnancy loss by 54% in women with APS and who are aPL-positive.

Mak *et al.* conducted a meta-analysis of studies involving women with RPL who were aPL-positive.[44] The study (N = 334) was conducted in order to ascertain whether the combination of heparin and LDA worked better than LDA alone to achieve live births. The authors found that the overall frequency of live births was 74.27% in women who used the combination compared to 55.83% in the group using LDA alone. Women who received a combination of heparin and LDA had a significantly higher live birth rate (RR = 1.301; 95% CI: 1.040, 1.629) than women who used LDA alone. The authors concluded that the combination of heparin and LDA resulted in more live births in aPL-positive women than using LDA alone.

In Cohn *et al's* study (N = 693), only 176 women (25%) were aPL-positive, while the rest had unexplained RPL.[45] Of the aPL-positive women, 69% (n = 122) had a subsequent live birth compared with 63% (n = 324) of the women with unexplained RPL (odds ratio [OR] = 1.3; 95% CI: 0.9–1.9). When the authors analysed the results, they found that 79% of aPL-positive women receiving a combination of heparin and LDA had a live birth compared with 62% of those receiving LDA alone (adjusted OR = 2.7; 95% CI: 1.3–5.8). In the unexplained RPL group, there was no difference in outcome for women receiving a combination of heparin and LDA or LDA only. The authors concluded that a combined

**Table 1:** Live birth rates in women with antiphospholipid syndrome and recurrent pregnancy loss treated with low-molecular-weight heparin plus low-dose aspirin compared with those treated with low-dose aspirin alone

| Study | Number of patients | Medication commencement in gestational weeks | Treatment for each group | Live birth rate % | *P* value |
|---|---|---|---|---|---|
| Empson *et al.* (2005)[43] | 119 | 7 | 1. LMWH + LDA<br>2. LDA | 73<br>34 | <0.05 |
| Mak *et al.* (2010)[44] | 334 | 6 | 1. LMWH + LDA<br>2. LDA | 74.27<br>55.83 | <0.05 |
| Cohn *et al.* (2010)[45] | 176 | 6 | 1. LMWH + LDA<br>2. LDA | 79<br>62 | <0.05 |
| Al Abri *et al.* (2000)[46] | 88 | 7 | 1. LMWH + LDA<br>2. LDA | 75<br>54 | <0.05 |

*LMWH = low-molecular-weight heparin; LDA = low-dose aspirin.*

treatment of LDA and heparin is superior to LDA alone in aPL-positive women, but not for women with unexplained RPL.

In their study, Al Abri *et al.* analysed the outcomes of pregnancies in a cohort of 21 Arab women and four women from other parts of Asia, who had had one or more episodes of fetal loss associated with raised levels of anticardiolipin antibodies.[46] They found that the rate of pregnancy loss was significantly higher in the first trimester than in the second or third. In the group that had received both LDA and prednisolone, 75% of pregnancies were successful compared to 54% in the group receiving aspirin alone, while 17% were successful in those who received no therapy. They concluded that LDA, either alone or with prednisolone, appears to improve significantly the chances for successful pregnancies in patients with anticardiolipin antibodies. The results of these studies are summarised in Table 1.

## Which Heparin should be used to Treat Antiphospholipid Syndrome and Antiphospholipid Antibodies?

One question to consider is which heparin should be used in women with APS and who are positive for aPL. In Fouda *et al.*'s study, 60 women with a history of three or more consecutive pregnancy losses and positive for aPL were divided equally into two groups.[47] One group received unfractionated heparin (5,000 units, twice daily) plus LDA. The second group received LMWH (enoxaparin 40 mg, once daily) plus LDA immediately after their pregnancy was confirmed. The authors found that 25 of the women who received LMWH (80%) and 20 of the women who received unfractionated heparin (66.67%) had successful pregnancies and deliveries ($P$ = 0.243). The authors concluded that LMWH plus LDA was a better alternative to unfractionated heparin plus LDA. Another advantage of LMWH is that it can be given once daily subcutaneously and can be self-administered.

## Is Inherited Thrombophilia Associated with Recurrent Pregnancy Loss?

The following studies, all of them prospective case-control studies, did not find an association between IT and adverse pregnancy outcomes.

In Said *et al.*'s prospective cohort study, 2,034 nulliparous women were recruited before 22 weeks' gestation.[48] Genotyping for FVL mutations, mutations of the prothrombin gene, methylenetetrahydrofolate reductase enzymes (MTHFR) C677T, MTHFR A1298C and thrombomodulin polymorphisms were performed. The thrombophilia investigation results were disclosed neither to the women nor to their physicians. The thrombophilia tests results and pregnancy outcomes were available in 1,707 women. Pregnancy complications such as placental abruption, severe pre-eclampsia, intrauterine growth restriction, stillbirths and early neonatal deaths occurred in 136 women (8%). The authors concluded that the majority of asymptomatic women with IT will have a successful pregnancy outcome.

Silver *et al.*'s study tried to ascertain whether women carrying mutations of the prothrombin gene *G20210A* were at higher risk of RPL, placental abruption, severe pre-eclampsia or intrauterine growth restriction.[49] They recruited 5,188 women, and 4,167 blood samples were taken in the first trimester and analysed for the gene mutation *G20210A*. In this study, only 3.8% of the women tested had a mutation of prothrombin *G20210A* and their pregnancy loss rates were similar to those of women without the mutation. The authors thus concluded that the prothrombin gene mutation *G20210A* was not associated with pregnancy loss or the other obstetric complications studied.

Dizon-Townson *et al.* studied pregnancy rates among women who were heterozygous carriers of the *FVL* mutation.[50] Only women with singleton pregnancies before 14 weeks' gestation were enrolled. In this study, the researchers failed to show any increase in pregnancy loss or other obstetric complications. such as pre-eclampsia, placental abruption or intrauterine growth restriction, when compared with non-carriers. They concluded that women who are heterozygous carriers of the *FVL* gene mutation do not require either screening or

treatment during pregnancy if there is no history of thromboembolisms.

Roqué *et al.* studied the association between inherited and acquired maternal thrombophilias and adverse pregnancy events.[51] A cohort of 491 patients with a history of adverse pregnancy outcomes was evaluated for activated protein C resistance, *FVL* and prothrombin *G20210A* mutations, hyperhomocysteinaemia, antithrombin deficiencies, proteins C and S, and both anticardiolipin antibodies and lupus anticoagulants. They found that the presence of maternal thrombophilia was not associated with an increased risk of fetal loss before 14 weeks' gestation.

Prospective case-control studies that showed an association between IT and adverse pregnancy outcomes were also found, and are summarised below.

In Preston *et al.*'s study, 1,384 women were enrolled in the European Prospective Cohort on Thrombophilia.[52] They found an increased risk of fetal loss in women with IT (168/571 *versus* 93/395; OR = 1.35; 95% CI: 1.01–1.82). The OR was greater for stillbirths than for miscarriages (3.6 [1.4–9.4] *versus* 1.27 [0.94–1.71]). The highest OR for stillbirth was in women with combined defects at 14.3 (2.4–86.0), which can be compared with 5.2 (1.5–18.1) in those with antithrombin deficiencies; 2.3 (0.6–8.3) in those with protein C deficiencies; 3.3 (1.0–11.3) in those with protein S deficiencies, and 2.0 (0.5–7.7) in those with *FVL* mutations. The OR for miscarriage in these subgroups was 0.8 (0.2–3.6) in women with combined defects; 1.7 (1.0–2.8) in those with antithrombin deficiencies; 1.4 (0.9–2.2) in those with protein C deficiencies; 1.2 (0.7–1.9) in those with protein-S deficiencies, and 0.9 (0.5–1.5) in women with *FVL* mutations. The authors concluded that women with familial thrombophilia, especially those with combined defects or antithrombin deficiencies, have an increased risk of fetal loss.

Kocher *et al.* studied 5,000 pregnant women and found a significant association between *FVL* mutations and stillbirths (OR = 19.9; 95% CI: 2.07–56.94), but not early fetal loss (OR = 1.76; 95% CI: 0.85–3.65).[53]

Sottilotta *et al.*'s study included 102 consecutive women with pregnancy loss who were tested for thrombophilia. Of these, 55 women with RPL (47 of whom had had a stillbirth) were tested for thrombophilia.[54] They found that the prevalence of prothrombin II, *FVL* and prothrombin *G20210A* mutations was higher in women with unexplained stillbirths. This finding was statistically significant. The prevalence of IT thrombophilia was higher in women with RPL, but the difference was not statistically insignificant.

## Treating Recurrent Pregnancy Loss in Cases of Inherited Thrombophilia

There are a few studies that show that treating women who suffer from IT and RPL with anticoagulants is beneficial; however, these studies have many limitations.

The LIVE-ENOX study compared two doses of LMWH (enoxaparin, 40 *versus* 80 mg) for women with and without thrombophilia.[55] Treatment with LMWH commenced between 5–10 weeks' gestation. The prevalence of thrombophilia was similar in both treatment groups and the live birth rate was 84.3% in women who received 40 mg of enoxaparin and 78.3% in women who received 80 mg of enoxaparin. Live birth rates were 70.0%, 84.4%, 76.9% and 81.3% for women with *FVL* mutations, hyperhomocysteinaemia, APS and other types of thrombophilia, respectively. The differences in these live birth rates were not significant (*P* = 0.484). The major limitation of this study was that the researchers did not compare LMWH with either LDA or a placebo.

In Deligiannidis *et al.*'s study, all of the subjects had RM and IT. The women in the experimental group (n = 29) received a combination of LMWH and LDA, while the control group (n = 23) received no treatment.[56] There was a significantly lower miscarriage rate among the women who received LMWH plus LDA when compared with those who received no treatment. However, the study was not randomised.

In Carp *et al.*'s cohort study, 58 women who had RM and IT were investigated.[57] Within this cohort, 37 received 40 mg of enoxaparin and 48 received no treatment. They found that 26 out of 37 (70.2%) of the women who received LMWH had live births compared with 21 out of 48 (43.8%) untreated women. (*P* <0.02; OR = 3.03; 95% CI: 1.12–8.36). The limitation of this study was that it was neither controlled nor randomised.

**Table 2:** Pregnancy outcomes in women with idiopathic recurrent pregnancy loss treated with low-molecular-weight heparin, prednisone, aspirin and a placebo

| Study | Number of patients | Medication commencement in gestational weeks | Treatment | Outcomes | | P value |
|---|---|---|---|---|---|---|
| | | | | Early miscarriage at <13 weeks | Late miscarriage at >13 weeks | |
| Badawy *et al.* (2008)[58] | 340 | 7 | 1. LMWH | 4.1 | 1.2 | <0.05 |
| | | | 2. Folic acid alone | 8.1 | 2.3 | |
| | | | | Live birth rate % | | |
| Fawzy *et al.* (2008)[59] | 160 | 7 | 1. LMWH | 81 | | <0.05 |
| | | | 2. Prednisone + LDA + progesterone | 85 | | |
| | | | 3. Placebo | 48 | | |

*LMWH = low-molecular-weight heparin; LDA = low-dose aspirin.*

# Treating Unexplained Recurrent Pregnancy Loss with Heparin

The use of LMWH in the treatment of patients with RPL without identified thrombophilia is based on two retrospective studies that reported a higher rate of successful pregnancy outcomes. Both studies had methodological limitations.

In Badawy *et al.*'s prospective randomised study, 340 women with first-trimester RM with no identifiable cause were investigated.[58] The women were randomised into two groups, with one group receiving LMWH and folic acid while the other group received only folic acid. In the LMWH group, treatment commenced once the fetal heartbeat was visible by ultrasound scan and continued until 34 weeks' gestation. Folic acid was discontinued at 13 weeks' gestation in both groups. The researchers found that the early miscarriage rate was 4.1% in the combined LMWH and folic acid group as compared with 8.8% in the folic acid alone group, while late pregnancy loss was 1.1% in the combined group compared with 2.3% in the folic acid alone group. This study showed modest improvements in the rates of pregnancy loss in women with RPL in the first trimester without thrombophilia (live birth risk ratio = 1.07; 95% CI: 1.00–1.14).

In Fawzy *et al.*'s prospective randomised, single-blinded, placebo-controlled study, 160 women diagnosed with idiopathic RM (more than three miscarriages) were recruited.[59] They received either LMWH (enoxaparin) alone; prednisone, LDA and progesterone, or a placebo in treatment, once the pregnancy had been confirmed by an ultrasound scan. Live birth rates were 85% in the combined therapy group and 81% in the LMWH alone group compared to 48% in women who received the placebo ($P$ <0.05).

The results of these studies are summarised in Table 2 and all show a favourable outcome in women with RPL, either with or without thrombophila.

# Prescribing Anticoagulants to Women with Recurrent Pregnancy Loss: Is the current practice justified?

Because of the results of the previously summarised investigations, the majority of clinicians worldwide have started prescribing heparin and/or aspirin for pregnant women with RPL, either with or without thrombophilia. However, because of the limitations of these studies, many researchers have conducted controlled, randomised, double-blind, multicentre studies to find out whether the current practice of prescribing anticoagulants to women with RPL with or without thrombophilia is justified. Those studies and their results are summarised below [Table 3].

The Scottish Pregnancy Intervention (SPIN) study was a multicentre, randomised, controlled trial of LMWH and LDA in women with RM.[60] This study included 294 women at less than seven weeks' gestation. All of the women had had two or more consecutive pregnancy losses before 24 weeks' gestation. Other causes of RPL were excluded, such as RPL due to endocrinological, chromosomal, immunological or anatomical abnormalities.

**Table 3:** A review of controlled, randomised, double-blind, multicentre studies to determine whether prescribing anticoagulants to women with recurrent pregnancy loss, with or without thrombophilia, is justified

| Trial | Number of patients | Miscarriages n (%) | Medication commencement in gestational weeks | Treatment for each group | Live birth rate % | P value |
|---|---|---|---|---|---|---|
| ALIFE (2010)[61] | 299 | 2 (40.1) ≥3 (59.9) | <6 | 1. LMWH + LDA 2. LDA 3. Placebo | 69 62 67 | 0.63 |
| HABENOX (2011)[7] | 207 | 2 (1.0) ≥3 (99.0) | <7 | 1. LMWH + LDA 2. LDA 3. LMWH | 65 61 71 | 0.45 |
| SPIN (2010)[60] | 294 | 2 (57.1) ≥3 (42.9) | <7 | 1. LMWH + LDA 2. Placebo | 78 80 | 0.85 |
| HepASA (2009)[62] | 88 | 2 (100) | <6 | 1. LMWH + LDA 2. LDA | 77.8 79.2 | 0.75 |

*ALIFE = Anticoagulants for Living Fetuses; LMWH = low-molecular-weight heparin; LDA = low-dose aspirin; HABENOX = Low Molecular Weight Heparin and/or Aspirin in Prevention of Habitual Abortion; SPIN = Scottish Pregnancy Intervention; HepASA = Heparin and Aspirin.*

Women were randomised into two groups. Group one received LMWH and LDA (40 mg of enoxaparin and 75 mg of aspirin daily) until 36 weeks' gestation with intensive surveillance, while group 2 had only intensive surveillance during the pregnancy. After randomisation, blood was sent from all of the women for thrombophilia testing. The results were not disclosed, either to the patient or to the clinician, until six weeks after the delivery. In this study, 10 women had IT, eight were heterozygous carriers of FVL mutations and five were identified in the pharmacological intervention group. Two cases had the prothrombin *G20210A* mutation, with one identified in each arm of the trial. The authors found that the pregnancy loss rate was 22% in women receiving both LMWH and LDA, while the pregnancy loss rate was 20% in the group who were only intensively monitored. They concluded that their results do not support the use of LMWH and/or LDA in women with RPL not due to APS.

In Kaandorp *et al.*'s randomised Anticoagulants for Living Fetuses (ALIFE) trial, the participants (n = 364) all had a history of unexplained RPL.[61] They were randomised equally into three groups. Group 1 received a daily dose of 80 mg of aspirin plus LMWH (2,850 IU) daily, group 2 received a daily dose of 80 mg of aspirin only and group 3 received a placebo. In this study, 299 women became pregnant. The live birth rates were similar in the three groups, with 69.1% (67/97) in group 1, 6% (61/99) in group 2 and 67.% (69/103) in group 3. This study showed that neither a combination of LMWH and LDA nor LDA alone improved the live birth rate when compared with the use of a placebo in women with RPL.

In Visser *et al.*'s Low Molecular Weight Heparin and/or Aspirin in Prevention of Habitual Abortion (HABENOX) study, a randomised, double-blind, multicentre study, 207 women were recruited.[7] All had had three or more consecutive miscarriages in the first trimester (less than 13 weeks' gestation), or two or more second trimester consecutive miscarriages. All women were tested for thrombophilia and were randomised before seven weeks' gestation to receive either LMWH (40 mg of enoxaparin) plus a placebo (n = 68), LMWH (40 mg of enoxaparin) plus 100 mg of aspirin (n = 63) or 100 mg of aspirin alone (n = 76). The live birth rate was 71% in women who received LMWH plus a placebo, 65% for those who received LMWH plus LDA and 61.5% in those who received LDA alone. The differences in the live birth rates were not statistically significant among the three groups, regardless of their thrombophilic statuses.

The Heparin and Aspirin (HepASA) trial studied 88 pregnant women with RPL who were either aPL-positive, or who had IT or antinuclear antibodies.[62] Women were randomised into two groups, with group one (n = 45) receiving LMWH plus LDA while group two (n = 43) received LDA alone. In each group, 47.7% of women were aPL-positive. The live birth rate was 77.8% (35/45) in group 1 and 79.1% (34/43) in group 2. The authors concluded that LMWH plus LDA showed no benefit over LDA alone in women with RPL.

## Conclusion

In women with RPL associated with IT, LMWH therapy has been shown to improve live birth rates when compared to LDA or a placebo. However, LMWH for women with RPL which is not associated with APS it is not recommended. In women with RPL and APS, LMWH can be used as early as six weeks' gestation until 34–36 weeks' gestation.

## References

1. Regan L, Rai R . Epidemiology and the medical causes of miscarriage. Baillieres Best Pract Res Clin Obstet Gynaecol 2000; 14:839–54.

2. Wilcox AJ, Weinberg CR, O'Connor JF, Baird DD, Schlatterer JP, Canfield RE, et al. Incidence of early loss of pregnancy. N Engl J Med 1988; 319:189–94.

3. Saravelos SH, Li TC. Unexplained recurrent miscarriage: How can we explain it? Hum Reprod 2012; 27:1882–6.

4. Hogge WA, Byrnes AL, Lanasa MC, Surti U. The clinical use of karyotyping spontaneous abortions. Am J Obstet Gynecol 2003; 189:397–400.

5. Sarig G, Younis JS, Hoffman R, Lamir N, Blumenfeld Z, Brenner B. Thrombophilia is common in women with idiopathic pregnancy loss and is associated with late pregnancy wastage. Fertil Steril 2002; 77:342–7.

6. Brenner B, Sarig G, Weiner Z, Younis J, Blumenfeld Z, Lanir N. Thrombophilic polymorphisms are common in women with fetal loss without apparent cause. Thromb Haemost 1999; 82:6–9.

7. Visser J, Ulander VM, Helmerhorst FM, Lampinen K, Morin-Papunen L, Bloemenkamp KWM, et al. Thromboprophylaxis for recurrent miscarriage in women with or without thrombophilia HABENOX*: A randomized multicentre trial. Thromb Haemost 2011; 105:295–301.

8. Colman-Brochu S. Deep vein thrombosis in pregnancy. MCN Am J Matern Child Nurs 2004; 29:186–92.

9. Abbate R, Lenti M, Fatini C, Gazzini A, Lapini I, Fedi S. L'ipercoagulabilità gravidica e puerperale. Haematologica 2003; 88:1–2.

10. Hathaway WE, Goodnight SH Jr. Thrombosis in pregnancy. In: Hathaway WE, Goodnight SH Jr, Eds. Disorders of Hemostasis and Thrombosis: A clinical guide, New York: McGraw-Hill Professional, 1993. Pp. 430–6.

11. de Boer K, ten Cate JW, Sturk A, Borm JJ, Treffers PE. Enhanced thrombin generation in normal and hypertensive pregnancy. Am J Obstet Gynecol 1989; 160:95–100.

12. Smyth A, Garovic VD. Systemic lupus erythematosus and pregnancy. Minerva Urol Nefrol 2009; 61:457–74.

13. Kaprara A, Krassas GE. Thyroid autoimmunity and miscarriage. Hormones (Athens) 2008; 7:294–302.

14. Castori M. Diabetic embryopathy: A developmental perspective from fertilization to adulthood. Mol Syndromol 2013; 4:74–86.

15. Akbaş H, Isi H, Oral D, Türkyılmaz A, Kalkanlı-Taş S, Simşek S, et al. Chromosome heteromorphisms are more frequent in couples with recurrent abortions. Genet Mol Res 2012; 11:3847–51.

16. Sanson BJ, Friederich PW, Simioni P, Zanardi S, Hilsman MV, Girolami A, et al. The risk of abortion and stillbirth in antithrombin-, protein C-, and protein S-deficient women. Thromb Haemost 1996; 75:387–8.

17. Finan RR, Tamim H, Ameen G, Sharida HE, Rashid M, Almawi WY. Prevalence of factor V G1691A (factor V-Leiden) and prothrombin G20210A gene mutations in a recurrent miscarriage population. Am J Hematol 2002; 71:300–5.

18. Lockwood CJ. Inherited thrombophilias in pregnant patients: Detection and treatment paradigm. Obstet Gynecol 2002; 99:333–41.

19. Sehirali S, Inal MM, Yildirim Y, Balim Z, Kosova B, Karamizrak T, et al. Prothrombin G20210A mutation in cases with recurrent miscarriage: A study of the Mediterranean population. Arch Gynecol Obstet 2005; 273:170–3.

20. Cosmi B, Legnani C, Pengo V, Ghirarduzzi A, Testa S, Poli D, et al. The influence of factor V Leiden and G20210A prothrombin mutation on the presence of residual vein obstruction after idiopathic deep-vein thrombosis of the lower limbs. Thromb Haemost 2013; 109:510–6.

21. Yildiz G, Yavuzcan A, Yildiz P, Süer N, Tandoğan N. Inherited thrombophilia with recurrent pregnancy loss in Turkish women--A real phenomenon. Ginekol Pol 2012; 83:598–603.

22. Nair RR, Khanna A, Singh K. MTHFR C677T polymorphism and recurrent early pregnancy loss risk in north Indian population. Reprod Sci 2012; 19:210–5.

23. Govindaiah V, Naushad SM, Prabhakara K, Krishna PC, Radha Rama Devi A. Association of parental hyperhomocysteinemia and C677T Methylene tetrahydrofolate reductase (MTHFR) polymorphism with recurrent pregnancy loss. Clin Biochem 2009; 42:380–6.

24. Azem F, Many A, Ben Ami I, Yovel I, Amit A, Lessing JB, et al. Increased rates of thrombophilia in women with repeated IVF failures. Hum Reprod 2004; 19:368–70.

25. Baumann K, Beuter-Winkler P, Hackethal A, Strowitzki T, Toth B, Bohlmann MK. Maternal factor V leiden and prothrombin mutations do not seem to contribute to the occurrence of two or more than two consecutive miscarriages in Caucasian patients. Am J Reprod Immunol 2013; 70:518–21.

26. Hoteit R, Taher A, Nassar R, Otrock Z, Halawi R, Mahfouz RA. Frequency of triple mutations involving factor V, prothrombin, and methylenetetrahydrofolate reductase genes among patients referred for molecular thrombophilia workup in a tertiary care center in Lebanon. Genet Test Mol Biomarkers 2012; 16:223–5.

27. Mukhopadhyay R, Saraswathy KN, Ghosh PK. MTHFR C677T and factor V Leiden in recurrent pregnancy loss: A study among an endogamous group in North India. Genet Test Mol Biomarkers 2009; 13:861–5.

28. McNeil HP, Simpson RJ, Chesterman CN, Krilis SA. Anti-phospholipid antibodies are directed against a complex antigen that includes a lipid-binding inhibitor of coagulation: Beta 2-glycoprotein I (apolipoprotein H). Proc Natl Acad Sci U S A 1990; 87:4120–4.

29. Ruiz-Irastorza G, Khamashta MA. [Systematic lupus erythematosus and antiphospholipid syndrome during pregnancy]. Z Rheumatol 2006; 65:192–4, 196–9.

30. Hauser AC, Hauser L, Pabinger-Fasching I, Quehenberger P, Derfler K, Hörl WH. The course of anticardiolipin antibody levels under immunoadsorption therapy. Am J Kidney Dis 2005; 46:446–54.

31. Hossain N, Shamsi T, Soomro N. Frequency of thrombophilia in patients with adverse pregnancy outcome. J Pak Med Assoc 2005; 55:245–7.

32. Molad Y, Borkowski T, Monselise A, Ben-Haroush A, Sulkes J, Hod M, et al. Maternal and fetal outcome of lupus pregnancy: A prospective study of 29 pregnancies. Lupus 2005; 14:145–51.

33. Levy RA, Avvad E, Oliveira J, Porto LC. Placental pathology in antiphospholipid syndrome. Lupus 1998; 7:S81–5.

34. Martínez-Zamora MÁ, Cervera R, Balasch J. Recurrent miscarriage, antiphospholipid antibodies and the risk of thromboembolic disease. Clin Rev Allergy Immunol 2012; 43:265–74.

35. Ganapathy R, Whitley GS, Cartwright JE, Dash PR, Thilaganathan B. Effect of heparin and fractionated heparin on trophoblast invasion. Hum Reprod 2007; 22:2523–7.

36. Ozdemir O, Yenicesu GI, Silan F, Köksal B, Atik S, Ozen F, et al. Recurrent pregnancy loss and its relation to combined parental thrombophilic gene mutations. Genet Test Mol Biomarkers 2012; 16:279–86.

37. Ivanov P, Komsa-Penkova R, Konova E, Gecheva S, Ivanov I, Kovacheva K, et al. [Combined thrombophilic factors among women with late recurrent spontaneous abortions]. Akush Ginekol (Sofiia) 2011; 50:8–12.

38. Hirsh J, Anand SS, Halperin JL, Fuster V. Mechanism of action and pharmacology of unfractionated heparin. Arterioscler Thromb Vasc Biol 2001; 21:1094–6.

39. Di Simone N, Caliandro D, Castellani R, Ferrazzani S, De Carolis S, Caruso A. Low-molecular weight heparin restores in-vitro trophoblast invasiveness and differentiation in presence of immunoglobulin G fractions obtained from patients with antiphospholipid syndrome. Hum Reprod 1999; 14:489–95.

40. Erden O, Imir A, Guvenal T, Muslehiddinoglu A, Arici S, Cetin M, et al. Investigation of the effects of heparin and low molecular weight heparin on E-cadherin and laminin expression in rat pregnancy by immunohistochemistry. Hum Reprod 2006; 21:3014–8.

41. Girardi G, Redecha P, Salmon JE. Heparin prevents antiphospholipid antibody-induced fetal loss by inhibiting complement activation. Nat Med 2004; 10:1222–6.

42. Han CS, Mulla MJ, Brosens JJ, Chamley LW, Paidas MJ, Lockwood CJ, et al. Aspirin and heparin effect on basal and antiphospholipid antibody modulation of trophoblast function. Obstet Gynecol 2011; 118:1021–8.

43. Empson M, Lassere M, Craig J, Scott J. Prevention of recurrent miscarriage for women with antiphospholipid antibody or lupus anticoagulant. Cochrane Database Syst Rev 2005; 2:CD002859.

44. Mak A, Cheung MW, Cheak AA, Ho RC. Combination of heparin and aspirin is superior to aspirin alone in enhancing live births in patients with recurrent pregnancy loss and positive anti-phospholipid antibodies: A meta-analysis of randomized controlled trials and meta-regression. Rheumatol (Oxford) 2010; 49:281–8.

45. Cohn DM, Goddijn M, Middeldorp S, Korevaar JC, Dawood F, Farquharson RG. Recurrent miscarriage and antiphospholipid antibodies: Prognosis of subsequent pregnancy. J Thromb Haemost 2010; 8:2208–13.

46. Al Abri S, Vaclavinkova V, Richens ER. Outcome of pregnancy in patients possessing anticardiolipin antibodies. J Sci Res Med Sci 2000; 2:91–5.

47. Fouda UM, Sayed AM, Abdou AM, Ramadan DI, Fouda IM, Zaki MM. Enoxaparin versus unfractionated heparin in the management of recurrent abortion secondary to antiphospholipid syndrome. Int J Gynaecol Obstet 2011; 112:211–5.

48. Said JM, Higgins JR, Moses EK, Walker SP, Borg AJ, Monagle PT, et al. Inherited thrombophilia polymorphisms and pregnancy outcomes in nulliparous women. Obstet Gynecol 2010; 115:5–13.

49. Silver RM, Zhao Y, Spong CY, Sibai B, Wendel G Jr, Wenstrom K, et al. Prothrombin gene G20210A mutation and obstetric complications. Obstet Gynecol 2010; 115:14–20.

50. Dizon-Townson D, Miller C, Sibai B, Spong CY, Thom E, Wendel G Jr, et al. The relationship of the factor V Leiden mutation and pregnancy outcomes for mother and fetus. Obstet Gynecol 2005; 106:517–24.

51. Roqué H, Paidas MJ, Funai EF, Kuczynski E, Lockwood CJ. Maternal thrombophilias are not associated with early pregnancy loss. Thromb Haemost 2004; 91:290–5.

52. Preston FE, Rosendaal FR, Walker ID, Briët E, Berntorp E, Conard J, et al. Increased fetal loss in women with heritable thrombophilia. Lancet 1996; 348:913–6.

53. Kocher O, Cirovic C, Malynn E, Rowland CM, Bare LA, Young BA, et al. Obstetric complications in patients with hereditary thrombophilia identified using the LCx microparticle enzyme immunoassay: A controlled study of 5,000 patients. Am J Clin Pathol 2007; 127:68–75.

54. Sottilotta G, Oriana V, Latella C, Luise F, Piromalli A, Ramirez F, et al. Genetic prothrombotic risk factors in women with unexplained pregnancy loss. Thromb Res 2006; 117:681–4.

55. Brenner B, Hoffman R, Carp H, Dulitsky M, Younis J, LIVE-ENOX Investigators. Efficacy and safety of two doses of enoxaparin in women with thrombophilia and recurrent pregnancy loss: The LIVE-ENOX study. J Thromb Haemost 2005; 3:227–9.

56. Deligiannidis A, Parapanissiou E, Mavridis P, Tabakoudis G, Mavroudi A, Papastavrou T, et al. Thrombophilia and antithrombotic therapy in women with recurrent spontaneous abortions. J Reprod Med 2007; 52:499–502.

57. Carp H, Dolitzky M, Inbal A. Thromboprophylaxis improves the live birth rate in women with consecutive recurrent miscarriages and hereditary thrombophilia. J Thromb Haemost 2003; 1:433–8.

58. Badawy AM, Khiary M, Sherif LS, Hassan M, Ragab A, Abdelall I. Low-molecular weight heparin in patients with recurrent early miscarriages of unknown aetiology. J Obstet Gynaecol 2008; 28:280–4.

59. Fawzy M, Shokeir T, El-Tatongy M, Warda O, El-Refaiey AA, Mosbah A. Treatment options and pregnancy outcome in women with idiopathic recurrent miscarriage: A randomized placebo-controlled study. Arch Gynecol Obstet 2008; 278:33–8.

60. Clark P, Walker ID, Langhorne P, Crichton L, Thomson A, Greaves M, et al. SPIN (Scottish Pregnancy Intervention) study: A multicenter, randomized controlled trial of low-molecular-weight heparin and low dose aspirin in women with recurrent miscarriage. Blood 2010; 115:4162–7.

61. Kaandorp SP, Goddijn M, van der Post JA, Hutten BA, Verhoeve HR, Hamulyák K, et al. Aspirin plus heparin or aspirin alone in women with recurrent miscarriage. N Engl J Med 2010; 362:1586–96.

62. Laskin CA, Spitzer KA, Clark CA, Crowther MR, Ginsberg JS, Hawker GA, et al. Low molecular weight heparin and aspirin for recurrent pregnancy loss: Results from the randomized, controlled HepASA trial. J Rheumatol 2009; 36:279–87.