Multiple thrombophilic single nucleotide polymorphisms lack a significant effect on outcomes in fresh IVF cycles: an analysis of 1717 patients

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Abstract

Purpose The aim of the study is to determine if thrombophilic single nucleotide polymorphisms (SNPs) affect outcomes in fresh in vitro fertilization (IVF) cycles in a large general infertility population.

Methods A prospective cohort analysis was performed at a university-affiliated private IVF center of female patients undergoing fresh non-donor IVF cycles. The effect of the following thrombophilic SNPs on IVF outcomes were explored: factor V (Leiden and H1299R), prothrombin (G20210A), factor XIII (V34L), β -fibrinogen (-455G→A), plasminogen activator inhibitor-1 (4G/5G), human platelet antigen-1 (a/b9L33P), and methylenetetrahydrofolate reductase (C677T and A1298C). The main outcome measures

included positive pregnancy test, clinical pregnancy, embryo implantation, live birth, and pregnancy loss.

Results Patients (1717) were enrolled in the study, and a total of 4169 embryos were transferred. There were no statistically significant differences in positive pregnancy test, clinical pregnancy, embryo implantation, live birth, or pregnancy loss in the analysis of 1717 patients attempting their first cycle of IVF. Receiver operator characteristics and logistic regression analyses showed that outcomes cannot be predicted by the cumulative number of thrombophilic mutations present in the patient.

Conclusions Individual and cumulative thrombophilic SNPs do not affect IVF outcomes. Therefore, initial screening for these SNPs is not indicated.

Keywords SNP · IVF outcomes · Thrombophilia · Infertility · Pregnancy

Clinical Trial Number (ClinicalTrials.gov): NCT01223092

Capsule Nine thrombophilic single nucleotide polymorphisms lack a significant effect on positive pregnancy test, clinical pregnancy, embryo implantation, pregnancy loss, and live birth in fresh non-donor IVF cycles.

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Introduction

Implantation limits the success of any given in vitro fertilization (IVF) cycle. Unfortunately, the complex interactions of endometrial and embryological factors that affect implantation are largely unknown. Implicated causes of unexplained infertility and recurrent pregnancy loss include inherited thrombophilias [1–3]. Disagreement exists regarding the effect inherited thrombophilias have on implantation.

Multiple studies support the belief that thrombophilias lead to recurrent implantation failure [4–7]. Specifically, a small study of patients with at least three failed IVF cycles found that factor V Leiden mutations and prothrombin G20210A mutations were more prevalent in the recurrent implantation failure group versus the fertile control group

[4]. Another study found the presence of any of the following inherited thrombophilias: prothrombin G20210A, factor V Leiden, methylenetetrahydrofolate reductase (MTHFR), protein S deficiency, protein C deficiency, or antithrombin III deficiency, was more common in recurrent implantation failure patients, especially if they had unexplained infertility, when compared with fertile controls [5]. A similar study testing the same set of inherited thrombophilias showed combinations of two or more of these thrombophilic mutations that were found more often in the recurrent implantation failure group when compared with controls of patients succeeding with their first IVF attempt or fertile patients that achieved pregnancy by natural conception [6]. When the thrombophilic panel was extended to ten thrombophilic single nucleotide polymorphisms (SNPs), patients with recurrent implantation failure, compared to fertile controls, showed a higher prevalence of the plasminogen activator inhibitor-1 (PAI-1) 4G/5G mutation and total number of inherited thrombophilic mutations [7]. To further support the role of the coagulation/fibrinolytic system in implantation, a study found reduced plasma fibrinolytic potential, demonstrated by increased clot lysis time, in patients experiencing recurrent implantation failure when compared with patients succeeding following their first cycle of IVF or fertile controls [8].

Conversely, multiple studies also exist that refute the idea of inherited thrombophilias affecting IVF outcomes [9–11]. These three studies examined the effects of the following inherited thrombophilias: factor V Leiden, prothrombin G20210A, and MTHFR. There were no differences found between infertility patients that failed IVF cycles versus fertile controls that conceived naturally. To complicate matters further, some studies found that the factor V Leiden mutation increased implantation [12, 13] while others found a decrease in first trimester miscarriage, increase in second trimester miscarriage, but otherwise no difference in fecundity [14]. Additionally, a study of MTHFR mutations in Brazilian women undergoing IVF due to male factor infertility found a difference in oocyte recovery and maturity but no difference in clinical pregnancy [15].

There is clearly a significant amount of disagreement in the literature regarding the role of inherited thrombophilias in implantation failure and IVF outcomes. The discordant findings between retrospective studies and small prospective studies has made it difficult for a meta-analysis to produce conclusive results [16]. This study attempts to resolve the disagreement in the literature by increasing power through more patients available for prospective analysis. It also attempts to make the results more applicable to the infertility specialist's practice by limiting the conclusions to comparisons made within the patient population seeking IVF for treatment of infertility.

Materials and methods

Study design and population

We prospectively enrolled 1717 female study patients undergoing their first fresh non-donor IVF cycle at a university-affiliated private IVF center from January 2006 to October 2010. No exclusion criteria were set, so all patients who chose to participate in the study that were undergoing their first fresh IVF cycle were included in this study. Patients provided follicular fluid or whole blood for DNA extraction prior to initiating IVF cycles to maintain a prospective study design. An independent institutional review board (IRB) approved this study.

We studied the following nine thrombophilic SNPs on IVF outcomes: factor V Leiden, factor V H1299R, prothrombin G20210A, factor XIII V34L, β -fibrinogen -455G→A, PAI-1 4G/5G, human platelet antigen-1 (HPA-1) a/b9L33P, MTHFR C677T, and MTHFR A1298C. Relevant IVF outcomes were positive pregnancy test, clinical pregnancy, embryo implantation, live birth, and pregnancy loss. For each of these outcomes, the patients with successful outcomes were compared to those with unsuccessful outcomes to determine if any of the thrombophilic SNPs could explain the differences. Counting the total number of thrombophilic mutations in the nine SNPs for each patient assessed the cumulative effect on IVF outcomes. SNP frequencies in the study population were compared against SNP frequencies in the HapMap CEU population, which is comprised of Utah residents with ancestry from Northern and Western Europe [17].

A serum hCG greater than 5 mIU/mL at 16 days following oocyte retrieval defined a positive pregnancy test. Evidence of at least one gestational sac by transvaginal ultrasound at 5–6 week gestational age defined a clinical pregnancy. The number of gestational sacs observed during the clinical pregnancy evaluation defined the number of implantation events. Embryo implantation rate was calculated for each patient by dividing the number of implantation events by the number of embryos transferred. We obtained the live birth data by patient/physician correspondence or follow-up phone calls to the patients as necessary. Subsequently, pregnancy loss was calculated both for patients who had a positive pregnancy test and for those who had a clinical pregnancy that did not result in a live birth.

Genotyping

DNA was isolated from follicular fluid or whole blood following the manufacturer's recommendations using the QIAmp DNA Blood BioRobot MDx Kit (Qiagen, Valencia, CA) on an epMotion 5075 VAC automated liquid handling instrument (Eppendorf, Westbury, NY). A

Nanodrop 8000 Spectrophotometer (Nanodrop Inc., Wilmington, DE) was used to quantify the DNA. In addition, agarose gel electrophoresis was used to characterize the integrity and size of each genomic DNA sample (1 % E-Gel; Invitrogen, Carlsbad, CA). PCR was performed for each DNA sample in duplicate with a custom or commercially available TaqMan SNP Genotyping Assay (Applied Biosystems Inc., Foster City, CA). The commercially available TaqMan SNP Genotyping Assays were prothrombin G20210A (Assay ID=C___8726802_20, dbSNP ID=rs1799963), factor V Leiden (Assay ID=C___11975250_10, dbSNP ID=rs6025), factor V H1299R (Assay ID=C___8919442_20, dbSNP ID=rs4525), MTHFR C677T (Assay ID=C___1202883_20, dbSNP=rs1801133), and MTHFR A1298C (Assay ID=C___850486_20, dbSNP ID=rs1801131). The custom TaqMan SNP Genotyping Assays for factor XIII V34L (dbSNP ID=rs5985), β -fibrinogen -455G $\rightarrow$ A (dbSNP ID=rs1800790), HPA-1 a/b9L33P (dbSNP=rs5918), and PAI-1 4G/5G (dbSNP ID=rs1799768) were generated with File Builder Version 3.1 (Applied Biosystems, Inc.) using the probes and primers shown in Supplemental Table 1. These assays were run in a GeneAmp 9700 Thermalcycler (Applied Biosystems Inc.) following the manufacturer's instructions. The genotype of each patient's DNA sample was determined using the allelic discrimination function within the SDS 2.3 analysis software (Applied Biosystems Inc.) running on a 7900HT Fast Real-Time PCR System (Applied Biosystems Inc.). The fluorescence quantified by the 7900HT was only used for input to the allelic discrimination software and thus will not be reported in this article.

Statistical analysis

We determined the differences between groups using Fisher's exact test (two-sided) for frequency data and the Kruskal-Wallis non-parametric one-way analysis of variance (ANOVA) test for determining differences between groups with respect to nominal variables. Categorical IVF outcomes were modeled with multivariate logistic regression. Receiver operator characteristics were generated for categorical IVF outcomes with respect to the cumulative number of thrombophilic mutations a patient possesses. "R" version 2.13.0 (<http://www.r-project.org/>) was the statistical software package used for performing all statistical calculations. The definition of *overall* experimental statistical significance was $\alpha < 0.05$ and was corrected for multiple comparisons secondary to the nine thrombophilic SNPs with the Bonferroni correction. The new corrected definition of statistical significance for any individual test was $\alpha_c < 0.0056$.

Results

There were 1717 patients (4169 embryos) attempting their first fresh cycle of IVF. Genotype data was unavailable for 2 patients for factor XIII V34L, 9 patients for factor V Leiden, 3 patients for factor V H1299R, 22 patients for β -fibrinogen -455G $\rightarrow$ A, 3 patients for MTHFR C677T, 5 patients for MTHFR A1298C, and 3 patients for PAI-1 4G/5G. Table 1 reports the demographic and stimulation parameters of the patients. There were 279 patients that received anticoagulation medication during their treatment cycle as follows: baby aspirin alone (253 patients), low molecular weight heparin with baby aspirin (14 patients), low molecular weight heparin with full dose aspirin (1 patient), and low molecular weight heparin alone (11 patients).

The following notation will be used for describing the genotypes of all SNPs: homozygous major as "AA," heterozygous as "Aa," and homozygous minor as "aa." Comparison of the means for the demographic and stimulation parameters across each genotype of the thrombophilic SNPs was statistically significant at a level α_c for height only with respect to the factor XIII V34L SNP (AA 64.2 in, Aa 64.5 in, aa 65.1 in, $P = 0.002$). Otherwise, there were no statistically significant differences in any of the demographic and stimulation parameters across each genotype for all other thrombophilic SNPs.

Table 2 shows the genotype frequencies and IVF outcomes. There were no statistically significant differences found in positive pregnancy test, clinical pregnancy, live birth, or embryo implantation at a level α_c . Combining the genotypes with the major allele for HPA-1 a/b9L33P and comparing it to the homozygous minor genotype was statistically insignificant at

Table 1 Patient demographics and stimulation parameters

Number of patients	1717
Mean patient age (years)	35.0 $\pm$ 4.4
Mean patient height (in)	64.3 $\pm$ 2.7
Mean patient weight (lbs)	149 $\pm$ 36.8
Mean patient BMI (kg/m ²)	25.4 $\pm$ 6.0
Mean partner age (years)	37.1 $\pm$ 5.7
Basal antral follicle count	14.1 $\pm$ 8.2
Max FSH (mIU/mL)	8.2 $\pm$ 3.2
Total gonadotropins administered (IU)	3045 $\pm$ 1282
No. of oocytes retrieved	14.7 $\pm$ 9.2
No. of blastocyst stage embryos	5.2 $\pm$ 4.1
No. of embryos transferred	2.4 $\pm$ 1.0
Peak estradiol (pg/mL)	1902 $\pm$ 1108
Peak endometrial thickness (mm)	10.0 $\pm$ 2.3

Values presented as mean $\pm$ standard deviation. Max FSH defines the maximum (day 3) basal FSH value measured at our institution. Number of blastocyst stage embryos is for day 5. Peak estradiol and peak endometrial thickness is on day of hCG administration

Table 2 IVF outcomes

SNP	Genotype frequency			Positive pregnancy test			Clinical pregnancy			Live birth			Embryo Implantation						
	AA	Aa	aa	AA	Aa	aa	P	AA	Aa	aa	P	AA	Aa	aa	P	AA	Aa	aa	P
Factor XIII V34L	63.2 %	32.2 %	4.6 %	62 %	66 %	52 %	0.042	53 %	57 %	44 %	0.086	43 %	44 %	33 %	0.187	35 %	37 %	28 %	0.115
	97.1 %	2.9 %	0 %	63 %	56 %	—	0.373	54 %	44 %	—	0.195	43 %	34 %	—	0.246	36 %	30 %	—	0.307
Prothrombin G20210A																			
Factor V Leiden	96.6 %	3.4 %	0 %	63 %	66 %	—	0.681	54 %	49 %	—	0.508	43 %	44 %	—	0.893	35 %	34 %	—	0.632
Factor V H1299R	55.3 %	37.8 %	6.9 %	62 %	63 %	65 %	0.850	53 %	55 %	55 %	0.558	42 %	45 %	42 %	0.392	35 %	37 %	35 %	0.588
β-fibrinogen -455G→A	65.0 %	31.2 %	3.8 %	62 %	63 %	55 %	0.404	53 %	56 %	42 %	0.105	42 %	45 %	33 %	0.137	35 %	36 %	28 %	0.224
HPHA-1 a/b9L33P	75.2 %	22.8 %	2.0 %	64 %	58 %	48 %	0.021	56 %	49 %	40 %	0.021	44 %	39 %	37 %	0.159	36 %	34 %	23 %	0.034
MTTHR C677T	46.5 %	41.4 %	12.1 %	60 %	65 %	65 %	0.103	52 %	56 %	55 %	0.227	41 %	45 %	44 %	0.250	33 %	37 %	37 %	0.135
MTTHR A1298C	46.0 %	43.8 %	10.2 %	63 %	62 %	61 %	0.866	53 %	55 %	51 %	0.677	43 %	43 %	41 %	0.801	35 %	36 %	33 %	0.707
PAI-1 4G/5G	26.8 %	49.2 %	24.0 %	64 %	62 %	64 %	0.680	55 %	53 %	56 %	0.524	42 %	42 %	44 %	0.863	36 %	34 %	38 %	0.408

The P values for positive pregnancy test, clinical pregnancy, and live birth were calculated using Fisher's exact test, while the P values for embryo implantation were calculated with Kruskal-Wallis ANOVA. All P -values refer to the data to the left of the value. To account for the Bonferroni correction, tests were considered statistically significant when $P < 0.0056$.

a level α_c for positive pregnancy test (AA/Aa 63 %, aa 48 %, $P=0.110$), clinical pregnancy (AA/Aa 54 %, aa 40 %, $P=0.122$), live birth (AA/Aa 43 %, aa 37 %, $P=0.605$), and embryo implantation (AA/Aa 36 %, aa 23 %, $P=0.034$). Furthermore, combining the genotypes with the minor allele for HPA-1 a/b9L33P and comparing it to the homozygous major genotype was statistically insignificant at a level α_c for positive pregnancy test (AA 64 %, Aa/aa 60 %, $P=0.013$), clinical pregnancy (AA 56 %, Aa/aa 48 %, $P=0.010$), live birth (AA 44 %, Aa/aa 39 %, $P=0.063$), and embryo implantation (AA 36 %, Aa/aa 33 %, $P=0.046$). There were no statistically significant differences in outcomes at a level α_c when treating the SNPs as recessive effects or dominant effects. Table 3 shows the pregnancy loss with respect to initial positive pregnancy test and clinical pregnancy for each thrombophilic SNP. There were no statistically significant differences in pregnancy loss at a level α_c .

The mean number of thrombophilic alleles per patient was 5.08. There was no statistically significant effect of the total number of thrombophilic mutations per patient on the odds of positive pregnancy test, clinical pregnancy, live birth, positive pregnancy test loss, and clinical pregnancy loss found with the logistic regression models after controlling for patient age and embryos transferred. Extending the logistic regression models to include if the patient received anticoagulation medication during treatment (including an interaction term with the number of thrombophilic alleles) does not reveal a statistically significant effect of anticoagulation on the outcomes ($P > 0.110$ for all variables except age and number of embryos transferred across all outcomes). Figure 1 shows the receiver operator characteristics for the categorical outcomes of positive pregnancy test, clinical pregnancy, live birth, positive pregnancy test loss, and clinical pregnancy loss by varying the cumulative number of thrombophilic mutations. All curves have an area under the curve (AUC) of approximately 0.5 and are almost identical to the random guess curve. Therefore, no threshold for the number of thrombophilic mutations exists that performs better than random prediction of IVF outcome.

There were no statistically significant differences between the HapMap CEU SNP genotype frequencies and this study group's frequencies (see Supplemental Table 2). HapMap data was not available for the prothrombin G20210A, β -fibrinogen -455G $\rightarrow$ A, and PAI-1 4G/5G SNPs.

Discussion

No differences were found for the IVF outcomes of positive pregnancy test, clinical pregnancy, embryo implantation, live birth, positive pregnancy test loss, or clinical pregnancy loss for any of the nine thrombophilic SNPs tested in this study.

Table 3 Pregnancy loss rates for each SNP

SNP	Positive pregnancy test loss				Clinical pregnancy loss			
	AA	Aa	aa	P	AA	Aa	aa	P
Factor XIII V34L	30 %	34 %	36 %	0.433	19 %	23 %	26 %	0.225
Prothrombin G20210A	32 %	39 %	–	0.413	20 %	23 %	–	0.790
Factor V Leiden	32 %	33 %	–	0.862	21 %	10 %	–	0.241
Factor V H1299R	34 %	28 %	35 %	0.185	21 %	19 %	23 %	0.570
β -fibrinogen -455G→A	33 %	29 %	40 %	0.227	21 %	19 %	22 %	0.794
HPA-1 a/b9L33P	32 %	33 %	24 %	0.728	21 %	21 %	7 %	0.567
MTHFR C677T	32 %	31 %	32 %	0.892	21 %	20 %	21 %	0.872
MTHFR A1298C	32 %	30 %	34 %	0.728	20 %	21 %	21 %	0.956
PAI-1 4G/5G	33 %	31 %	31 %	0.758	23 %	19 %	21 %	0.545

All *P* values were calculated using Fisher's exact test and refer to the data to the left of the *P* value

Furthermore, no threshold exists for the cumulative number of thrombophilic mutations a patient possesses that can assist in predicting IVF outcomes. Therefore, screening for the presence of these nine thrombophilic SNPs does not assist the infertility specialist in the initial diagnostic workup of the patient. Screening prior to the first cycle of IVF would only increase the cost of care.

There were no statistically significant differences at the level α_c found for any of the IVF outcomes when examining the nine SNPs individually. It is tempting to interpret the *P*

values for the frequency data which were less than the uncorrected α as statistically significant, but this would not be statistically correct given that nine SNPs were simultaneously analyzed against the same outcome data. If the study used one independent group for each SNP, then comparing the *P* values to an uncorrected α would have been appropriate.

We can calculate the power of the statistical calculations for each SNP to detect a *clinically significant* difference to determine if the study had sufficient statistical power to detect such a difference. Assuming a clinically significant difference to be

Fig. 1 Receiver operator characteristics for IVF outcomes. *AUC* area under the curve. Sensitivity and specificity are functions of the cumulative number of thrombophilic mutations a patient possesses

at least 10 % between the SNP genotypes while assuring the Bonferroni $\alpha_c=0.0056$ with the genotype frequencies shown in Table 2, the power for each SNP to detect a difference in live birth (and embryo implantation) is as follows: factor XIII V34L 84 % (89 %), prothrombin G20210A 9 % (10 %), factor V Leiden 10 % (12 %), factor V H1299R 87 % (92 %), β -fibrinogen -455G $\rightarrow$ A 83 % (87 %), human platelet antigen-1 a/b9L33P 72 % (77 %), MTHFR C677T 88 % (92 %), MTHFR A1298C 88 % (92 %), and plasminogen activator inhibitor-1 4G/5G 77 % (85 %), where the number in parenthesis represents the power of the embryo implantation statistic for that SNP. All SNPs except prothrombin G20210A and factor V Leiden achieve a power greater than 70 % to detect a clinically significant difference in live birth and embryo implantation even after accounting for the Bonferroni correction to assure an *overall* experimental α of 0.05. The decreased power from the prothrombin G20210A and factor V Leiden SNPs is secondary to the lower frequencies of these mutations. The low frequency decreases the effect size in the power calculation which subsequently requires more samples to achieve the same power when compared to the other SNPs.

The IVF outcomes of the HPA-1 SNP, the SNP with P values closest to reaching significance at a level α_c , revealed a dose response of positive pregnancy test and clinical pregnancy that was inversely related to increasing number of minor alleles. Embryo implantation appeared to be diminished only for the homozygous minor genotype. In order to increase statistical power for this calculation, we grouped the homozygous major genotype with the heterozygous genotype to reduce the degrees of freedom in the statistical calculation. This new group of patients that possessed at least one major allele was compared to the homozygous minor genotype, but it still failed to reach a level of α_c for statistical significance for the HPA-1 SNP. The HPA-1 minor allele is known to lower the threshold for platelet activation, alpha-granule release, and fibrinogen binding while increasing platelet sensitivity to antiplatelet drugs. [18, 19]. The impact of missing a true effect of the minor allele for HPA-1 is overlooking a potential medical intervention. For example, an antiplatelet drug administered to patients identified with the HPA-1 SNP could increase embryo implantation and thus pregnancy if the HPA-1 SNP truly affects implantation.

A statistically significant difference in implantation was not found for the PAI-1 4G/5G SNP that was demonstrated by Coulam et al. [7] in a study of recurrent implantation failure patients. Our study has sufficient power for the PAI-1 4G/5G SNP to detect a modest 10 % difference in embryo implantation, but we did not detect any statistically significant difference in this population. Our results also disagree on the negative impact of increasing number of thrombophilic SNPs a patient possesses found by Coulam et al. [7]. Our

receiver operator characteristics graphically show that there is no threshold for the number of thrombophilic SNPs a patient possesses that can reliably predict IVF outcomes. Furthermore, even after adjusting for patient age and number of embryos transferred with a logistic regression model, we still did not find a statistically significant effect of the number of thrombophilic mutations a patient possesses. An important difference between this study and the one by Coulam et al. [7] which may explain the discrepancy is that they used fertile controls for comparison whereas we used patients seeking IVF treatment as their own controls.

This study does not support increased embryo implantation with factor V Leiden mutation carriers previously described [12, 13]. The probability of a positive pregnancy test was higher for the factor V Leiden mutation carriers, but the difference was not statistically significant. We agree with the conclusion of Rudick et al. [13] that screening for factor V Leiden in the general IVF population is not indicated.

We found no statistically significant difference in pregnancy loss for the factor V Leiden or prothrombin G20210A mutation, but a meta-analysis of non-recurrent and recurrent pregnancy loss found these mutations to be associated with increased loss [3]. A possible explanation, in addition to the decreased statistical power of these SNPs in our study as explained above, for the discrepancy is that the population in our study was an infertility population whereas the populations pooled for the meta-analysis were not.

This study has multiple strengths. First, all patients were offered IRB-approved consent to use their DNA for research purposes. Although some patients refused to use their DNA for research purposes, this study included all the patients that consented to DNA use who were undergoing their first fresh IVF cycle. This makes the study results more applicable to the infertility specialist treating a patient considering IVF. Furthermore, by including only the first IVF cycle instead of subsequent failed cycles, the study results are more applicable to determining if these SNPs are useful as a screening test prior to initiating IVF cycles. Including patients that failed multiple cycles will also obscure any effects of the SNPs because other factors, such as poor egg quality, are likely more significant. Second, it utilizes a large prospective patient DNA database as the data source. Prospective data collection decreases the potential selection bias attributed to patients with prior poor outcomes being more willing to participate in a research study. Third, we collected live birth data, which is ultimately the outcome of interest to the patient. Finally, calculating embryo implantation rather than inferring successful implantation via positive pregnancy test allows for better documentation of a clinical pregnancy.

Despite the study's multiple strengths, it also has weaknesses. The simultaneous analysis of multiple thrombophilic SNPs on the same set of outcome data necessitates a more stringent requirement on P values to have an overall

experimental α level of statistical significance. The Bonferroni correction, which is a conservative correction for multiple statistical comparisons, was used to assure an overall experimental α of 0.05. This may have led to an increased number of false negatives. By reporting all P values regardless of statistical significance, the data generated by the study can be reanalyzed by using more liberal statistical methods for controlling for multiple comparisons.

In summary, the thrombophilic SNPs studied in this cohort of infertile patients undergoing fresh IVF cycles had no individual or cumulative effect on positive pregnancy test, clinical pregnancy, embryo implantation, live birth, or pregnancy loss. Given these results, it is unlikely that screening for common thrombophilic SNPs is indicated for the infertility specialist's initial diagnostic evaluation.

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Compliance with ethical standards

Conflict of interest The authors of this paper have no conflicts of interest, financial or otherwise, to declare.

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