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Prevalence of Parental Thrombophilic Defects After Fetal Death and Relation to Cause

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OBJECTIVE: To estimate whether parental thrombophilic defects after fetal death, either acquired or inherited, were more prevalent than in the normal population and to estimate associations between these thrombophilic defects and different fetal death causes.

METHODS: In a multicenter, prospective cohort study of 750 fetal deaths, we tested couples for antithrombin, protein C, total and free protein S, and von Willebrand factor (vWF) plasma levels. Mothers' values were compared with reference values in gestational age-matched healthy pregnant women, and fathers were compared with healthy men. Prevalence of factor V Leiden, prothrombin *G20210A* mutation, and lupus anticoagulant were compared with the normal population. A panel classified death cause.

RESULTS: More women with fetal death had decreased antithrombin (16.8%, *P*<.001) and protein C (4.0%, *P*=.03) and increased vWF (15.5%, *P*<.001) plasma levels than healthy pregnant women (2.5%). However, compared with normal ranges in the nonpregnant population, we only observed more women with increased vWF (12.4%, *P*<.001). More fathers had decreased free protein S (6.3%, *P*<.001) and elevated vWF (12.1%, *P*<.001) than healthy men (2.5%). Prevalence of inherited thrombophilias was not higher in couples with fetal death than in the population. Neither inherited nor acquired maternal or paternal thrombophilic defects were associated with the main cause of death. Of placental causes, abruption and infarction were associated with acquired maternal defects.

CONCLUSION: Except for vWF and paternal free protein S, acquired and inherited thrombophilic defects were not more prevalent after fetal death. Routine thrombophilia testing after fetal death is not advised.
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LEVEL OF EVIDENCE: II

Approximately one in 200 pregnancies ends in stillbirth as a result of different causes.[1] Maternally inherited thrombophilic defects are inconsistently recognized as risk factors for pregnancy complications such as preeclampsia, placental abruption, growth restriction, and stillbirth,[2,3] whereas paternal thrombophilic factors can also be transferred to the fetus and placenta.[4] Many studies have addressed the association between inherited thrombophilic deficiencies and late fetal loss in families with inherited deficiencies, whereas case–control and cohort studies have studied women with fetal loss who were tested for thrombophilia after delivery. In reviews, a higher risk for fetal loss was observed for antithrombin and protein S deficiency, factor V Leiden, the prothrombin *20210A* mutation, and anticardiolipin antibodies.[5–7]

The pathophysiology of late fetal loss associated with inherited thrombophilia is presumed to be placental thrombosis either in the maternal or fetal circulation of the placenta leading to placental infarction and placental insufficiency.[8] This hypothesis has been difficult to prove as a result of the small numbers

[†]*Deceased.*

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involved in previous studies, whereas placental insufficiency was often poorly defined and not related to cause of death.[9–12] Pregnancy is a hypercoagulable state resulting from acquired thrombophilic defects exposing women to a higher risk of thrombosis.[13] In combination with preexisting inherited thrombophilic defects, it may increase the risk of fetal death. Little is known about the contribution of acquired thrombophilic defects to the hypercoagulable state and fetal death because women in previous studies were only tested for thrombophilia several weeks after delivery.

The objective of our study was to estimate whether in a cohort of women with intrauterine fetal death, thrombophilic defects, either acquired during pregnancy or preexisting inherited, and inherited defects in fathers were more prevalent than in the normal population. Furthermore, we estimated the association between these thrombophilic defects and the various fetal death causes within our cohort.

MATERIALS AND METHODS

In 2002, we initiated a prospective intrauterine fetal death cohort study in 50 Dutch secondary and tertiary referral hospitals serving rural as well as urban populations. Inclusion criteria were singleton intrauterine fetal death diagnosed antepartum (heart beat stopped before labor) after 20 weeks of gestation calculated from the last menstrual period and confirmed by ultrasonography. Terminations were excluded. The study was approved by the review boards of all the participating hospitals, and written informed consent was obtained from participants. Data were collected for each intrauterine fetal death, including medical and obstetric history, maternal and fetal characteristics, and pregnancy and delivery details. Our diagnostic workup protocol was based on local protocols and included maternal blood tests, including full blood count, chemistry, and viral serology; coagulation tests for couples performed centrally at the laboratory in Groningen; fetal blood tests, including viral serology; microbiologic cultures from the mother, fetus, and placenta; autopsy; placental examination; and cytogenetic analysis.

In women with intrauterine fetal death, plasma levels of antithrombin, protein C activity, total and free protein S antigen, and von Willebrand factor (vWF) were measured in samples collected on induction of labor. These plasma levels were compared with plasma reference values in 110 healthy pregnant women of comparable gestational age recruited from our obstetrics department after informed consent was obtained. Women were excluded if they had an individual or family history of venous thromboembolism, known thrombophilia, complications in past pregnancies or the present one, or if they used medication. Healthy pregnant women were tested at four intervals of gestation: 12–16 weeks, 28–32 weeks, in an early stage of labor (greater than 34 weeks), and 5–7 weeks postpartum. Reference plasma values were determined for all cases for gestational age periods from 20–27 weeks, 27–34 weeks, and greater than 34 weeks (in an early stage of labor). Values at 20–27 weeks of gestation were obtained by linear estimation from values at 12–16 weeks and 28–32 weeks for each individual. Plasma levels of antithrombin, protein C activity, total protein S antigen, and free protein S antigen in women with intrauterine fetal death were defined as abnormal if they were below 2.5% of the values in healthy pregnant women at comparable gestational age or above 97.5% for vWF. Use of these cutoff values is common when determining reference values.

We also measured plasma levels of antithrombin, protein C activity, total protein S antigen, free protein S antigen, and vWF in the fathers and we determined normal plasma ranges in 393 healthy men recruited as blood donors in our hospital who had no (family) history of venous or arterial thromboembolism. Abnormal plasma levels were defined as levels below the lower limit of the normal ranges: antithrombin less than 74 international units/dL; protein C activity less than 64 international units/dL; total protein S antigen less than 67 international units/dL; and free protein S antigen less than 65 international units/dL. von Willibrand factor levels greater than 150 international units/dL were increased. All these abnormal plasma levels are referred to as "thrombophilic defects."

Antithrombin (Chromogenix, Mölndal, Sweden) and protein C activity (Dade Behring, Marburg, Germany) were measured by chromogenic substrate assays; total protein S antigen, free protein S antigen, and vWF were measured by enzyme-linked immunosorbent assay (DAKO, Glostrup, Denmark).

In men and women, inherited thrombophilias factor V Leiden and the prothrombin *G20210A* mutation were determined by polymerase chain reactions. Lupus anticoagulant was determined as previously described.[14] Prevalence of these thrombophilias was compared with prevalence in the normal population.

Autopsies and placental examinations (including histology) were performed by surgical as well as perinatal pathologists in the participating hospitals. We urged pathologists to follow the study protocol, which was based on the guidelines published by the Royal College of Obstetricians and Gynecologists

and the Royal College of Pathologists[15] and the College of American Pathologists.[16,17] Fetal growth percentiles for birth weight by gestational age at time of diagnosis of intrauterine fetal death were calculated according to Kloosterman's growth charts.[18] Small for gestational age was defined as birth weight less than the 10th percentile.

Cause of fetal death was classified by a multidisciplinary panel according to the Tulip classification,[19] which covers six main causes: congenital anomaly, placental pathology, prematurity or immaturity, infection, other (ie, maternal diseases, fetal hydrops), or unknown. The cause was classified as unknown if other causes had been excluded. Risk factors, such as smoking and preeclampsia, were defined as contributing to death.

Causes of deaths resulting from maternal and fetal placental circulation pathology were 1) placental abruption, a clinical diagnosis supported by placental examination; 2) significant infarction in preterm cases, any placental infarction, and in term cases, extensive infarction (greater than 10%) of the placental area. Infarction as a cause was allocated if the percentage of infarctions in the parenchyma in relation to the weight of the placenta was regarded likely to cause death. In a term placenta of appropriate weight, at least 30% infarctions was regarded plausible to cause death.[20] This is in accordance with others[21]; 3) fetal thrombotic vasculopathy, the presence of avascular villi (at least one focus of five or more villi), thrombosis in a vessel of the chorionic plate or stem villus, hemorrhagic endovasculitis, or intramural fibrin in a vessel of the chorionic plate or stem villus in the absence of umbilical cord blood flow restriction[22,23]; and 4) maternal floor infarct or massive perivillous fibrin deposition, extensive periv-

Table 1. Characteristics of the Study Population: Women

Thrombophilic Defect	None (n=447)	At Least One (n=267)	*P*
No.	62.6%	37.4%	
Family history			
Venous thromboembolism (first degree)	4.9	8.5	.07
Known hereditary thrombophilia	1.5	4.2	.06
Personal history			
Venous thromboembolism	0.9	4.9	.001
Known hereditary thrombophilia	0	2.0	.006
Previous IUFD	2.7	3.8	.50
Recurrent early fetal loss	6.3	5.6	.87
Age (y) [median (range)]	32 (18–46)	30 (18–46)	.005
Ethnic origin			
White*	88.4	86.1	.31
African American	2.7	5.2	
Eastern	4.3	3.7	
Other	4.7	4.9	
Pregnancy			
Nulliparous	49.7	56.9	.20
Primiparous	21.9	18.4	
Multiparous	28.4	24.7	
Hypertension-related disease†	11.0	25.8	<.001
Diabetes-related disease‡	4.1	3.4	.84
Smoking	23.3	25.9	.83
Anticoagulant thromboprophylaxis	1.6	4.5	.028
Current IUFD			
Gestational age (wk) [mean (SD)]	34.4 (6.3)	28.4 (5.7)	<.001
Birth weight (g) [median (range)]	2,000 (40–4,630)	810 (12–4,425)	<.001
Small for gestational age§	28.8	43.0	<.001
Time lag diagnosis and birth (d) [median (range)]	2 (0–40)	2 (0–23)	.27

IUFD, intrauterine fetal death; SD, standard deviation.
Coagulation tests were available in 714 (95.2%); results are given in percent unless otherwise indicated.
* Including Mediterranean groups.
† Hypertension-related disease (chronic hypertension; pregnancy-induced hypertension; preeclampsia; hemolysis, elevated liver enzymes, low platelet syndrome; and superimposed conditions) based on recommendations by the International Society for the Study of Hypertension in Pregnancy.
‡ Diabetes-related (type I diabetes and gestational diabetes).
§ According to Kloosterman's growth charts,[18] which start at 25 weeks of gestation.

illous fibrin deposition, either predominantly basally located or diffusely distributed in at least 30% of the parenchyma.[24]

Other categories of placental pathology were placental hypoplasia, an absolute too-low placenta weight (less than the 10th percentile), a too-low placenta or birth weight ratio, or both[25] and other placental pathology such as villus immaturity and umbilical cord complications.

Sample size of our initial national intrauterine fetal death study was based on the ability to identify less frequent but clinically relevant causes of death (ie, 5% rather than formal hypothesis testing). To achieve sufficient precision, we considered a lower boundary of the 95% confidence interval (CI) of 3.5%. This resulted in a sample size of 750 fetal deaths. Categorical variables were expressed as counts and percentages and continuous data as means with standard deviation or median and ranges with exact 95% CIs given when appropriate. Differences between groups for categorical data were evaluated by the Fisher's exact test or χ^2 test. For continuous variables, we used the Student's *t* test or Mann-Whitney *U* test, depending on the normality of data. A two-tailed $P<.05$ was considered to indicate statistical significance. Statistical analyses were performed using SAS software 9 (SAS Institute Inc, Cary, NC).

RESULTS

From 2002 to 2006, we enrolled 750 couples. Coagulation tests were available in 714 (95.2%) women and 664 (88.5%) men. Autopsies were performed in 525 (70.0%) fetal deaths and placental examinations in 736 (98.1%).

The characteristics of men and women with at least one thrombophilic defect (22.4% and 37.4%, respectively) and those without such defects are shown in Tables 1 and 2). A personal history of venous thromboembolism (4.9%) and known thrombophilia (2.0%), hypertension-related disease (25.8%), and anticoagulant thromboprophylaxis during pregnancy (4.5%) were observed more often in women with a thrombophilic defect. In this subgroup, median age at pregnancy, mean gestational age at delivery (28.4 weeks compared with 34.4 weeks), and median birth weight were lower, whereas it also contained more small-for-gestational-age babies.

Compared with reference values in nonpregnant women, the majority of women with intrauterine fetal death and healthy pregnant women had a tendency to higher antithrombin levels, no difference in protein C activity levels, a tendency to lower total protein S antigen, lower free protein S antigen, and higher vWF levels (Fig. 1). Compared with healthy pregnant women, women with intrauterine fetal death had lower levels of antithrombin and higher levels of vWF up to 34 weeks of gestation.

Regarding acquired thrombophilic defects during pregnancy, women with intrauterine fetal death more often had significantly decreased levels of antithrombin (16.8%) and protein C antigen (4.0%) and increased vWF levels (15.5%) compared with healthy pregnant women (2.5%; Tables 3 and 4). Decreased free protein S antigen levels were less common (0.9%). When compared with plasma levels in the normal, nonpregnant population, decreased levels of antithrombin (3.7%, $P=.07$) and protein C antigen (2.1%, $P=.62$) were not observed more often in contrast to increased vWF levels (87.6%, $P<.001$) that were still observed more often in women with intrauterine fetal death. The prevalence of preexisting inherited thrombophilias factor V Leiden

Table 2. Characteristics of the Study Population: Men

Thrombophilic Defect	None (n=515)	At Least One (n=149)	*P*
No.	77.6%	22.4%	
Family history			
Venous thromboembolism (first degree)	4.4	6.9	.26
Known hereditary thrombophilia	0.9	2.3	.19
Personal history			
Venous thromboembolism	0.4	0.7	.53
Known hereditary thrombophilia	0.2	0.8	.40
Age (y) [median (range)]	34 (18–61)	35 (19–60)	.01
Current IUFD			
Gestational age (wk) [mean (SD)]	31.6 (6.3)	31.7 (6.7)	.74
Birth weight (g) [median (range)]	1,485 (12–4,560)	1,395 (51–4,630)	.98
Small for gestational age*	33.5	35.3	.74

IUFD, intrauterine fetal death; SD, standard deviation.
Coagulation tests were available in 664 (88.5%) men; results are given in percent unless otherwise indicated.
* According to Kloosterman's growth charts,[18] which start at 25 weeks of gestation.

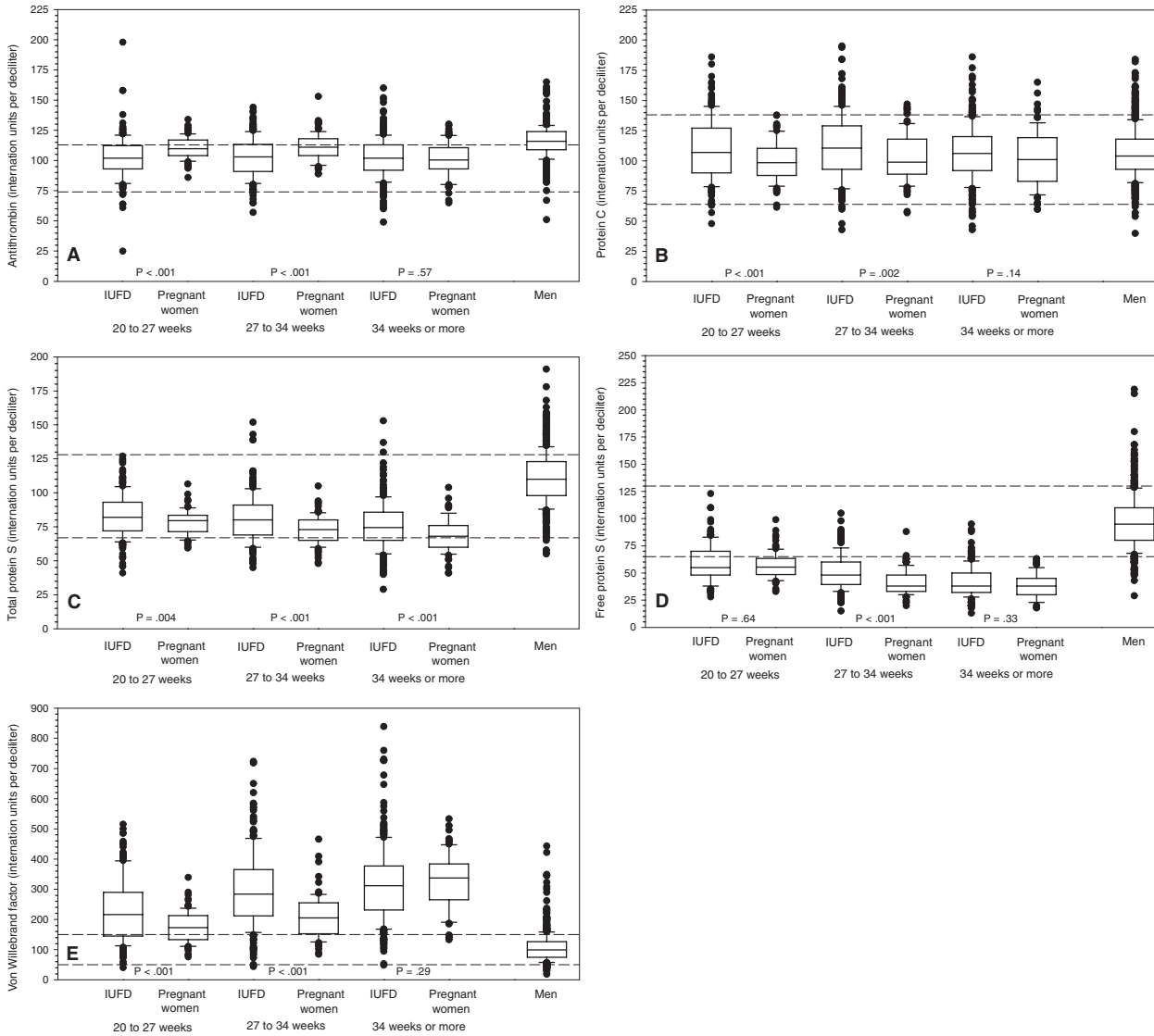

Fig. 1. Plasma levels of natural anticoagulant proteins (antithrombin, protein C, total protein S, free protein S) and von Willebrand factor according to gestational age (20–27 weeks, 27–34 weeks, and 34 weeks or more) in women with intrauterine fetal death (IUFD), healthy pregnant women (Pregnant women), and male partners of women with IUFD (Men) compared with reference values in the normal nonpregnant population (*dotted line*). *P* values indicate comparison between women with IUFD compared with healthy pregnant women. **A.** Antithrombin. **B.** Protein C. **C.** Total protein S. **D.** Free protein S. **E.** von Willebrand factor.

Korteweg. Thrombophilia Testing After Fetal Death. Obstet Gynecol 2010.

and prothrombin *G20210A* mutation in women with intrauterine fetal death was comparable to the normal population, whereas lupus anticoagulant (1.4%) was observed less frequently than expected. Men in the intrauterine fetal death group more often had decreased free protein S antigen plasma levels (6.3%) and elevated vWF levels (12.1%) compared with healthy men (2.5%).

Causes of death were placental pathology (64.9%), congenital anomaly (5.3%), infection (1.9%), other (4.8%), or unknown (23.1%: 15.9% despite thorough investigation and 7.2% resulting from insufficient information). Table 5 presents thrombophilic defects found in women with intrauterine fetal death (n=750) and their partners in relation to cause of death. A thrombophilic defect was seen in 39.1% of women with a placental cause compared with 34.1% with a nonplacental cause. Overall, in both men and women, none of the separate thrombophilic defects were associated with placental compared with nonplacental causes. Decreased maternal total protein S

Table 3. Prevalence of Thrombophilic Defects in Couples With Intrauterine Fetal Death

Plasma Levels Abnormal	Healthy Pregnant Women* (%)	Percent of Women IUFD (No. Tested [CI])	*P*	Healthy Men* (%)	Percent of Men IUFD (No. Tested [CI])	*P*
Antithrombin ↓	2.5	16.8 (702 [14.1–19.8])	<.001	2.5	0.3 (655 [0.04–1.1])	<.001
Protein C ↓	2.5	4.0 (708 [2.6–5.7])	.03	2.5	0.6 (661 [0.2–1.5])	<.001
Total protein S ↓	2.5	3.0 (707 [1.9–4.5])	.48	2.5	0.5 (659 [0.1–1.3])	<.001
Free protein S ↓†	2.5	0.9 (699 [0.3–1.9])	.003	2.5	6.3 (656 [4.5–8.4])	<.001
vWF ↑	2.5	15.5 (702 [12.9–18.4])	<.001	2.5	12.1 (659 [9.7–14.9])	<.001

IUFD, intrauterine fetal death; CI, confidence interval; vWF, von Willebrand factor.
* Reference values.
† Free protein S ↓ but normal total protein S.

antigen plasma levels were more often associated with nonplacental causes, and elevated maternal vWF levels were associated with "other cause of death." In addition, when we considered the 267 women with a thrombophilic defect, 182 (68%) had an intrauterine fetal death as a result of a placental cause (39.1% of the 465 women with a placental cause). This was comparable to the group of women without thrombophilic defects (283 of 447 [63%], *P*=.20). Of the 149 men with a thrombophilic defect, 94 (63%) were in the intrauterine fetal death group with placental causes (21.7% of 434 men overall with a placental cause), which is comparable to 340 of 515 men (66%, *P*=.56) without a defect. No association with placental causes was observed in couples in which there was both a maternal and a paternal thrombophilic defect (32 of 53 [60%] compared with 199 of 317 [63%], *P*=.19) compared with couples without a defect.

Analysis of placental causes of death showed that death was the result of infarction in 99 of 182 (54%) and abruption in 30 of 182 (16%) women with a thrombophilic defect (Table 6). Compared with all the different placental causes, abruption was more frequently associated with decreased levels of antithrombin (40.8%, *P*<.001), protein C activity (20.4%, *P*<.001), and total protein S antigen (10.2%, *P*<.001) and increased vWF levels (18.4%, *P*=.03); infarction was associated with decreased antithrombin (26.1%, *P*<.001) and elevated vWF (28.4%, *P*<.001) levels and lupus anticoagulant (2.4%, *P*=.04) and maternal floor infarct and massive perivillous fibrin deposition with elevated vWF (28.6%, *P*<.001). Abruption was seen significantly more often in women with abnormal plasma levels of antithrombin, protein C activity, and total protein S antigen compared with infarction. Overall, of placental causes, abruption and infarction were most frequently observed in women with thrombophilic defects (*P*<.001).

DISCUSSION

Our intrauterine fetal death cohort study was primarily set up to evaluate valuable diagnostics to determine cause of fetal death. We addressed the contribution of preexisting inherited as well as acquired thrombophilic defects during pregnancy to fetal death. Thrombophilia testing was therefore performed at induction of labor and protein levels in women with fetal death were compared with healthy pregnant women of comparable gestational age. We defined protein levels as potential risk factors for thrombosis in pregnancy (ie, as thrombophilic defects) when they were less than the 2.5th percentile in healthy pregnant women for antithrombin, protein C activity, and protein S antigen and greater than the

Table 4. Prevalence of Inherited Thrombophilic Defects in Couples With Intrauterine Fetal Death

	Prevalence in the Normal Population	Percent Women With IUFD (No. Tested [CI])	*P*	Prevalence in the Normal Population	Percent Men With IUFD (No. Tested [CI])	*P*
Factor V Leiden	5%	6.7 (689 [4.9–8.8])	.06	5%	4.2 (642 [2.8–6.1])	.41
Heterozygous		6.4			4.0	
Homozygous		0.3			0.2	
Prothrombin G20210A	3%	3.0 (691 [1.9–4.6])	1.0	3%	0.9 (642 [0.3–2.0])	<.001
Heterozygous		3.0			0.9	
Homozygous		—			—	
Lupus anticoagulant	3%	1.4 (646 [0.6–2.6])	.01	3%	0 (105 [0–3.5])	.08

IUFD, intrauterine fetal death; CI, confidence interval.

Table 5. Thrombophilic Defects in Couples With Intrauterine Fetal Death (N=750) in Relation to Cause of Death

Abnormal Plasma Levels	Placental Causes (n=487)	Nonplacental Causes					*P**
		Congenital (n=40)	Infection (n=14)	Other (n=36)	Unknown (n=173)	Total (n=263)	
Antithrombin ↓							
Women	18.5 (459)	13.9 (36)	21.4 (14)	18.8 (32)	11.8 (161)	13.6 (243)	.11
Men	0.5 (428)	0 (36)	0 (11)	0 (32)	0 (148)	0 (227)	.55
Protein C ↓							
Women	4.1 (462)	0 (38)	21.4 (14)	3.1 (32)	3.1 (162)	3.7 (246)	.84
Men	0.5 (432)	0 (36)	0 (12)	0 (32)	1.3 (149)	0.9 (229)	.61
Total protein S ↓							
Women	2.0 (461)	2.6 (38)	7.1 (14)	3.1 (32)	5.6 (162)	4.9 (246)	.04
Men	0.2 (430)	0 (36)	0 (12)	0 (32)	1.3 (149)	0.9 (229)	.28
Free protein S ↓[†]							
Women	0.9 (456)	0 (38)	0 (14)	0 (32)	1.3 (159)	0.8 (243)	>.99
Men	4.9 (430)	5.9 (34)	8.3 (12)	12.5 (32)	8.8 (148)	8.9 (226)	.06
vWF ↑							
Women	17.1 (457)	7.9 (38)	14.3 (14)	34.4 (32)	9.3 (161)	12.7 (245)	.13
Men	12.6 (429)	11.1 (36)	25.0 (12)	9.4 (32)	10.7 (150)	11.3 (230)	.71
Factor V Leiden							
Women	7.6 (447)	0 (37)	21.4 (14)	3.1 (33)	5.1 (158)	5.0 (242)	.20
Men	4.5 (419)	5.7 (35)	0 (12)	3.1 (32)	2.8 (144)	3.1 (223)	.41
Prothrombin *G20210A*							
Women	3.4 (448)	0 (37)	7.1 (14)	0 (33)	3.1 (159)	2.5 (243)	.65
Men	1.0 (419)	0 (37)	0 (12)	0 (32)	1.4 (144)	0.9 (223)	>.99
Lupus anticoagulant							
Women	1.0 (422)	3.0 (33)	7.1 (14)	6.7 (30)	0.7 (147)	2.2 (224)	.29
Men	NA						
Any defect							
Women	39.1 (465)	21.1 (38)	64.3 (14)	51.5 (33)	31.1 (164)	34.1 (249)	.20
Men	21.7 (434)	22.2 (36)	33.3 (12)	25.0 (32)	23.3 (150)	23.9 (230)	.56
Couples	7.4 (432)	5.6 (36)	16.7 (12)	12.5 (32)	8.7 (149)	9.1 (229)	.19

vWF, von Willebrand factor; NA, not applicable.
Percentages (no. tested) are given.
* *P* value for comparison of placental versus nonplacental causes.
[†] Free protein S ↓ but normal total protein S.

97.5th percentile for vWF. Testing for thrombophilia after fetal death may be useful in clinical practice if the results can be used to prevent recurrent fetal loss. Our data provide no support for routine testing of inherited or acquired thrombophilic defects after fetal death, although acquired defects during pregnancy may play a role in deaths caused by abruption or infarction.

Overall, in women with fetal death, levels of antithrombin and protein C antigen remained within the normal ranges for nonpregnant women. These levels are not related to a greater risk for thrombosis in nonpregnant women, but cutoff levels for fetal loss in pregnant women may differ. On the other hand, these proteins may contribute to fetal loss through mechanisms other than their anticoagulant properties, for example, cell protection, inhibition of apoptosis of trophoblast cells, and antiinflammatory effects.[26] This assumption was supported by our finding that intrauterine fetal death was diagnosed at earlier gestational age and with more small-for-gestational-age fetuses in women with these thrombophilic defects. However, the higher rate of hypertension-related disease in this group could also account for this with the thrombophilia being an epiphenomenon. Decreased levels of protein S antigen and increased levels of vWF in most healthy pregnant women, compared with reference values in nonpregnant women, also suggested different cutoff levels in pregnancy.

Significantly increased maternal vWF plasma levels were observed in our intrauterine fetal death group compared with healthy pregnant women in which vWF levels increase during normal pregnancy,[13] but it is unknown when these levels become pathologic. von Willibrand factor activity was found to be higher in women with early miscarriage than in

Table 6. Thrombophilic Defects in 487 Women With Intrauterine Fetal Death Resulting From Placental Pathology

Abnormal Plasma Levels	Abruption (n=52)	Infarction (n=197)	FTV (n=2)	MFI/MPFD (n=7)	Combination Group 1 to 4* (n=30)	Hypoplasia (n=90)	Other Placental (n=109)	P†
Antithrombin ↓	40.8 (49)	26.1 (184)	0 (2)	14.3 (7)	7.1 (28)	9.3 (86)	5.8 (103)	<.001
Protein C ↓	20.4 (49)	3.3 (184)	0 (2)	0 (7)	0 (30)	2.3 (87)	1.0 (103)	<.001
Total protein S ↓	10.2 (49)	1.7 (184)	0 (2)	0 (7)	0 (30)	1.2 (87)	0 (102)	.002
Free protein S ↓‡	4.1 (49)	0.6 (183)	0 (2)	0 (7)	0 (29)	0 (87)	1.0 (99)	.31
vWF ↑	18.4 (49)	28.4 (183)	0 (2)	28.6 (7)	10.7 (28)	12.8 (86)	1.0 (102)	<.001
Factor V Leiden	4.2 (48)	8.0 (176)	0 (2)	0 (7)	6.7 (28)	3.6 (83)	12.9 (101)	.28
Prothrombin *G20210A*	6.3 (48)	3.4 (177)	0 (2)	0 (7)	0 (30)	3.6 (83)	3.0 (101)	.85
Lupus anticoagulant	0 (48)	2.4 (167)	0 (1)	0 (6)	0 (26)	0 (79)	0 (95)	.41
Any defect women	61.2 (49)	52.9 (187)	0 (2)	42.9 (7)	23.3 (30)	25.3 (87)	20.4 (103)	<.001
Couples	15.2 (46)	9.8 (173)	0 (2)	0 (7)	0 (29)	5.1 (79)	4.2 (96)	.008

FTV, fetal thrombotic vasculopathy; MFI, maternal floor infarct; MPFD, massive perivillous fibrin deposition; vWF, von Willebrand factor.

Percentages (no. tested) are given.

* Combination groups 1 to 4: a combination of one of the following causes of death: abruption, infarction, FTV, and MFI/MPFD.

† *P* value indicates differences between all subgroups.

‡ Free protein S ↓ but normal total protein S.

control subjects.[27] We speculated that our results could be related to non-O blood type[28] or an acute phase response; non-O blood type was indeed associated with higher vWF levels, whereas C-reactive protein and fibrinogen were not (data not shown).

Abnormal paternal plasma levels of free protein S antigen and vWF were observed in the intrauterine fetal death group. Others reported no difference in fetal mortality in women with partners with and without inherited thrombophilia.[29] In contrast, a doubled prevalence (60%) of numerous thrombophilic defects in partners of women with a history of perinatal mortality compared with control participants (30%) was reported.[30]

Overall, factor V Leiden, prothrombin *G20210A* mutation, and lupus anticoagulant were not associated with fetal death. This is in contrast with earlier studies that reported relative risks for late fetal loss of 2.1–3.3 for maternal factor V Leiden, 2.3–3.0 for maternal prothrombin *G20210A* mutation, and 2.4 for maternal lupus anticoagulant.[6,7,31] Variation in population characteristics could explain these differences.

Trophoblast invasion of the maternal uterine circulation, spiral artery remodeling, and maintenance of blood fluidity in the intervillous space require a balance between prothrombotic and antithrombotic forces. Most deaths in our study were caused by placental pathology. Overall for main cause of death, none of the maternal and paternal thrombophilic defects were related to a placental cause nor for couples with double thrombophilic defects. However, analysis of the various placental causes of fetal death showed that thrombophilic defects were associated with placental abruption and infarction. Abruption was associated with decreased levels of antithrombin, protein C activity, and total protein S antigen and increased vWF levels; infarction with decreased antithrombin levels; increased vWF levels; and lupus anticoagulant. Furthermore, both pathologies were associated with combined thrombophilic defects and thrombophilic defects in fathers (decreased free protein S antigen, increased vWF levels). Because placentas were examined by different surgical as well as perinatal pathologists, we must presume that there is interobserver variation resulting in underestimation of placental pathologies that are relatively difficult to establish. Our results suggest that acquired thrombophilic defects may play a role in deaths caused by abruption or infarction, which represent one third of fetal deaths.

Measuring protein levels at the start of induction in women with fetal death might have influenced our results, because these were compared with healthy pregnant women at comparable gestational age. Such an effect seems less likely after comparing the results in the various subgroups. The median time between diagnosis of intrauterine fetal death and birth was 2 days, which makes it unlikely that dead fetus syndrome played a role.[32] Placental abruption might be the cause rather than the result of associated thrombophilic defects resulting from disseminated intravascular coagulation. Similarly, hypertension-related disease may cause changes in protein levels as a result of impaired liver function. To address possible con-

founding factors, we performed two extra subgroup analyses. First, we excluded women who might have had disseminated intravascular coagulation as a result of abruption and, second, women with hypertension-related disease. The results were similar to our overall analyses, indicating limited, if any, confounding (data not shown).

The need for routine testing of thrombophilic defects after fetal death is not supported by our results, except in women with a family history of hereditary thrombophilia or a personal history of venous thromboembolism and intrauterine fetal death, in whom testing could help prevent further maternal venous thromboembolisms.[5] Testing of women with a fetal death caused by abruption or infarction can be considered. However, thrombophilia screening should only be performed in cases in which proper management can be offered. As long as there are no randomized controlled trials proving the benefits of anticoagulant therapy in cases with known thrombophilic factors in relation to late fetal loss, we must be reserved about implementing a potentially harmful intervention in pregnant women. Testing for abnormal levels of antithrombin, protein C activity, total protein S antigen, or vWF may yield predictors for a subgroup at risk for fetal death caused by abruption or infarction.

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