

Protein C and S Deficiency, Thrombophilia, and Hypofibrinolysis: Pathophysiologic Causes of Legg-Perthes Disease

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ABSTRACT. In eight patients with Legg-Perthes disease, we assessed the etiologic roles of thrombophilia caused by protein C and protein S deficiency and hypofibrinolysis mediated by low levels of tissue plasminogen activator activity. We speculated that thrombosis or hypofibrinolysis were common causes of Legg-Perthes disease. Three of the eight patients had protein C deficiency; they came from kindreds with previously undiagnosed protein C deficiency. In one of these three kindreds there were six protein C-deficient family members (beyond the proband child), four of whom had thrombotic events as adults. One of the eight patients had protein S deficiency, as did his brother who had sustained mesenteric vein thrombosis at age 43. One of the eight patients who had normal proteins C, S, and antithrombin III had hypofibrinolysis, failing to elevate tissue plasminogen activator activity after 10 min of venous occlusion at 100 mm Hg. Plasminogen activator inhibitor, α_2 -antiplasmin, and fibrinogen values were normal in all eight patients. Beyond their Legg-Perthes disease, none of the eight patients had evidence for venous thrombosis. Of the eight patients, four had thrombophilia and one had hypofibrinolysis, disorders that we believe contributed to thrombotic venous occlusion of the femur with subsequent venous hypertension and bone death that characterize Legg-Perthes disease. (*Pediatr Res* 35: 383-388, 1994)

Abbreviations

PAI, plasminogen activator inhibitor
tPA, tissue plasminogen activator
tPA-Ag, tissue plasminogen activator antigen
tPA-Fx, tissue plasminogen activator activity
PAI-Fx, plasminogen activator inhibitor activity
PAI-Ag, plasminogen activator inhibitor antigen

We suspect that Legg-Perthes disease in children is a pediatric equivalent of idiopathic osteonecrosis of the femoral head in the adult (1-9). The cause of Perthes disease is unknown, and no uniform agreement exists regarding how best to treat the disease process (1-9). In dog models, when venous drainage of the surgical neck of the femur is obstructed, avascular necrosis resembling that of Legg-Perthes disease develops in the femoral head (8). It has been postulated that Legg-Perthes disease may

be caused by intravascular thrombosis as a result of reduced fibrinolysis (9).

We have recently shown that hypofibrinolysis mediated by high levels of the major inhibitor of fibrinolysis, PAI, is a common major cause of idiopathic osteonecrosis (10, 11). Nine of 12 adults with idiopathic osteonecrosis had high levels of PAI with hypofibrinolysis (10, 11). The thrombogenic, atherogenic apolipoprotein, Lp(a), was elevated in 14 of 18 patients with secondary osteonecrosis, and protein C deficiency was present in 1 of the 18 patients, suggesting that thrombophilia and hypofibrinolysis also play major pathophysiologic roles in secondary osteonecrosis (11).

Estimates of the prevalence of heterozygous protein C deficiency vary widely from less than 1 in 200 000 (12), to 1 in 16 000 (13), to 1 in 200 to 300 (14). Even among patients with venous thromboembolic disease, low protein C levels ranging from 35 to 65% of the normal mean (presumed heterozygous protein C deficiency) are uncommon, being found in eight of 319 (1 of 40) (13) and two of 225 (1 of 113) (15) cases.

In studies of Dutch patients with heterozygous protein C deficiency 50% of heterozygotes and 10% of normal relatives had venous thromboembolism by age 45 y, and 50% of first thromboembolic episodes and 65% of recurrences were spontaneous, not preceded by surgery or pregnancy (16). The authors concluded that "... members of the family of a symptomatic heterozygote proband who are heterozygous for the mutation in the protein C gene have an increased risk of venous thrombotic events compared with their normal family members." Homozygous protein C deficiency is rare and often fatal, seen in neonates with disseminated intravascular coagulation and purpura fulminans (16).

Heterozygous protein S deficiency appears to be rare even among patients with proven acute deep vein thrombosis, seen in only 2.8% of 107 patients with thrombophilia (17).

Until our recent studies (10, 11), only six kindreds with familial hypofibrinolysis resulting from high PAI had been reported. Hence, heritable, primary protein C, S, and antithrombin III deficiencies (12-17) are rare, and familial hypofibrinolysis resulting from high PAI is rare (10, 11).

Considerable variability exists in estimates of the prevalence of Perthes disease, ranging from 1 in 1 200 in Massachusetts (18), 1 in 1 400 in British Columbia (19), 1 in 4 750 in South Wales (20), and 1 in 12 500 in the United Kingdom (21).

Assuming the prevalence of heterozygous protein C deficiency to be as high as 1 in 250 (14) and the prevalence of Perthes disease to be as high as 1 in 1 300 (18, 19), then by chance alone the likelihood that a child would have Perthes disease and concurrent heterozygous protein C deficiency would, speculatively, be 1 in 325 000 ($1/250 \times 1/1 300$). Within this frame of reference, and because hypofibrinolysis and thrombophilia play

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a major role in adult idiopathic and secondary osteonecrosis (10, 11), our specific aim was to study the potentially etiologic roles of thrombophilia and hypofibrinolysis in eight patients who had Legg-Perthes disease.

MATERIALS AND METHODS

Patients. After identifying all cases with the diagnosis of Legg-Perthes disease by review of patient records in two large clinical orthopedic practices, we obtained the names of 14 patients for potential follow-up. Of these 14 patients, selected solely by diagnosis codes, 5 no longer lived in Cincinnati, and 1 refused to participate; 8 (57%) consented to have measures of fibrinolytic activity and quantitation of proteins C, S, and antithrombin III. No selection bias whatsoever existed. Because family and clinical histories were not obtained until the patients' first visit at our Cholesterol Center, we had no foreknowledge of other heritable disorders and no *a priori* postulates that could have affected patient selection in any way. All eight patients had typical age of presentation and characteristic clinical, radiologic, and physical findings of Legg-Perthes disease (1-9). Their osteonecrosis was not secondary to known causes such as corticosteroids, sickle cell disease, or hip fracture (1, 11). The work followed a research protocol approved by the Jewish Hospital Medical Research Council and was carried out with signed informed consent.

Measures of fibrinolytic activity, protein C, S, and antithrombin III. Basal and stimulated determinants of fibrinolysis and D-dimer were quantitated as previously reported (11, 22). After the patients fasted for 12-h, blood was obtained between 0800 h and 0900 h in seated patients to minimize the influence of time of day on regulators of basal and stimulated fibrinolytic activity (11). In seven of the eight probands, while they remained seated and after measurement of basal fibrinolytic regulators and D-dimer, stimulated fibrinolysis was measured after a standard stimulus (10, 11), 10 min of venous occlusion at 100 mm Hg with a blood pressure cuff. Although we obtained measures of fibrinogen, plasminogen, and α_2 -antiplasmin before and after venous occlusion (in addition to tPA, PAI, and D-dimer), we did not display the postocclusion values for fibrinogen, plasminogen, and α_2 -antiplasmin because they are primarily affected by hemoconcentration and are not released from the endothelium in response to increased vascular transmural pressure.

tPA, the major fibrinolytic activity stimulator, was measured in two forms, tPA-Ag by ELISA (Imubind total tPA, American Diagnostica, Greenwich, CT), and tPA-Fx by a chromogenic technique [Biopool Spectrolyse (fibrin), American Diagnostica] (11, 22).

PAI-Fx, a major inhibitor of fibrinolysis (10, 11, 22-24), was measured with synthetic substrates [Biopool Spectrolyse (pL), American Diagnostica] (11, 22). This assay measures only the functionally active PAI. PAI-Ag was quantitated with MAb and ELISA (Imubind PAI-1, American Diagnostica) (11, 22). PAI-Ag measures total antigen, *i.e.* both latent (bound to tPA) plus active PAI (free).

Additional measures of fibrinolytic regulators included α_2 -antiplasmin (a fibrinolytic inhibitor) (S-2251, KabiVitrum, Franklin, OH), plasminogen (Spectrozyme PL, American Diagnostica), and fibrinogen (Dade Thrombin, Baxter Healthcare Corp., Miami, FL) (11, 22).

D-dimers, split products from cross-linked fibrin, a measure of *in vivo* fibrinolysis (11, 22), were determined by ELISA (Dimer-test, American Diagnostica) with MAb against D-dimer.

Protein C was measured by ELISA (Imubind Protein C, Strip-Well, American Diagnostica).

Protein S and antithrombin III were measured respectively by Rellplate S, American Diagnostica, and Dade Antithrombin III chromogenic assay (Baxter Healthcare Corp) (25, 26). Free protein S was determined immunologically (25). In the proband with familial protein S deficiency, a functional assay for protein S was also used (27) (Diagnostica Stago). C4B binding protein

was also determined with an immunologic method (28) (American Bio Products, Parsippany, NJ).

Measurements of lipids, lipoprotein cholesterol, and apolipoproteins. Fasting lipids and lipoprotein cholesterol were quantitated in our Lipid Research Clinic's standardized laboratory by enzymatic methods (29-31), with calculation of LDL cholesterol when the triglyceride value was less than 400 mg/dL. Serum apolipoproteins A1 and B were measured by rate-nephelometry (32). Lp(a) was measured by enzyme immunoassay with a Terumo-macra Lp(a) sequential monoclonal-polyclonal sandwich method (33).

RESULTS

Probands. As summarized in Table 1, we studied eight patients, including the following: six children and two young adults, seven males and one female, seven white and one black. The two young adults had Legg-Perthes disease at ages 8 y (patient 3) and 6 y (patient 8). We also studied all available first-degree relatives. All eight patients had typical age of disease onset and characteristic clinical, radiologic, and physical findings of Legg-Perthes disease (1-9, 34).

Although two of the eight patients (patients 3 and 8, Table 1) were studied decades after their clinical presentation, from our experience with hereditary thrombophilia and hypofibrinolysis in adults with idiopathic osteonecrosis (10, 11) and from the literature on protein C and S deficiency (12-17, 27, 28), heritable hypofibrinolytic and thrombophilic defects are persistent from childhood, not transient.

Unexpectedly, and heretofore not previously reported (1-9, 34), four of the eight patients with Perthes had low LDL cholesterol values (patients 1, 5, 7, and 8; Table 1). Of these four patients with hypobetalipoproteinemia, patient 8 had familial protein S deficiency, patient 7 had protein C deficiency, and patient 5 could not initiate fibrinolysis (Table 1). Of the three probands with protein C deficiency (probands 2, 6, and 7; Table 1), proband 7 had familial hypobetalipoproteinemia (35) with very low total and LDL cholesterol and apo B; he also had Stickler's syndrome (36, 37), a pleiomorphic, dominantly transmitted trait that includes retinal detachment, degenerative arthritis, and, occasionally, cleft palate.

Proband 3 had moderately elevated total and LDL cholesterol, and proband 6 had elevated Lp(a) (Table 1).

Measures of basal and stimulated determinants of fibrinolysis, D-dimer, and proteins C, S, and antithrombin III. Overall, four of the eight probands had thrombophilic diseases [three with protein C deficiency (probands 2, 6, and 7) and one with protein S deficiency (proband 8; Table 1)]. One of the eight probands had hypofibrinolysis (proband 5).

Three of the probands (probands 2, 6, and 7) had previously undiagnosed low levels of protein C; their protein S and antithrombin III levels were normal (Table 1). Beyond their Legg-Perthes disease, these three probands with protein C deficiency had no personal history of thrombophlebitis or venous thrombosis.

Family studies in kindred 2 (Fig. 1, Table 2) revealed that the Legg-Perthes proband's mother, maternal grandfather, uncle, aunt, sister, and female first cousin all had protein C deficiency. All four adult family members with protein C deficiency had experienced venous thrombosis, pulmonary emboli, or both; none had Legg-Perthes disease or osteonecrosis (10, 11) (Fig. 1). The proband's maternal grandfather (I-1, Fig. 1) was on coumadin when sampled because of recurrent thrombosis, which further depressed his low protein C levels. The only thrombotic event in generation III was in the pediatric proband with Legg-Perthes disease (III-3), presumably affecting the venous drainage of the femur (Fig. 1).

In addition to familial, three-generation, autosomal dominant, protein C deficiency in kindred 2, there was also familial (10), three-generation, autosomal dominantly transmitted high (top

Table 1. Fibrinolytic, thrombogenic, and lipid characteristics of eight probands with Legg-Perthes disease

	Normal ranges	Proband/kindred*							
		1 (10, M, B)	2† (7, M, W)	3 (28, M, W)	4 (11, F, W)	5 (14, M, W)	6 (7, M, W)	7 (15, M, W)	8 (36, M, W)
Protein C (%)	55-145	84	48‡	91	64	64	41‡	54‡	85
Total protein S-Ag (%)	70-150	85	86	98	73	74	73	81	16§
Free protein S-Ag (%)	65-135	102	92	93	75	78	90	ND	<10§
Antithrombin III (%)	80-120	94	105	86	106	83	92	116	95
Basal tPA-Fx (IU/mL)	0.11-1.94	0.50	2.48	0.64	0.72	0.28	0.61	0.54	0.64
Stimulated tPA-Fx (IU/mL)	2.28-11.3	3.72	ND	12.85	3.60	0.34	3.87	2.87	0.64
Basal tPA-Ag (ng/mL)	3.7-13.7	4.60	7.20	9.20	7.10	5.0	11.4	6.8	8.1
Stimulated tPA-Ag (ng/mL)	8.4-31.4	11.7	ND	49.2	20.4	7.0	32.9	18.6	11.0
Basal PAI-Fx (U/mL)	3.5-26.9	5.6	6.4	9.8	14.6	6.8	18.8	5.5	4.2
Stimulated PAI-Fx (U/mL)	3.6-23.9	5.8	ND	0.50	14.6	6.2	16.2	6.2	1.6
Basal PAI-Ag (ng/mL)	3.2-37.1	4.6	16.0	19.6	22.4	10.1	35.1	16.2	ND
Stimulated PAI-Ag (ng/mL)	11.7-95.6	8.7	ND	26.1	26.0	11.0	43.0	18.8	ND
Basal α ₂ -antiplasmin (%)	77-129	118	117	106	106	93	102	102	115
Basal plasminogen (%)	75-133	91	94	56	80	77	74	121	90
Basal fibrinogen (mg/dL)	150-400	210	229	193	210	197	204	229	244
Basal D-dimer	<0.25	0.09	0.206	0.08	0.091	0.114	0.098	ND	ND
Stimulated D-dimer	0.085-0.257	0.092	ND	0.084	0.090	0.129	0.102	ND	ND
Cholesterol (mmol/L)	<5.17	3.08	4.60	5.43	3.85	3.44	3.70	2.22	3.98
Triglyceride (mmol/L)	<2.82	0.73	0.36	0.89	0.55	0.55	0.53	0.87	0.80
HDLc (mmol/L)	>0.91	1.19	1.99	1.40	1.47	1.55	1.47	0.80	1.16
LDLC (mmol/L)	<4.14	1.55¶	2.46	3.62	2.12	1.63¶	1.99	1.03¶	2.46
Lp(a) (mg/dL)	<20	1	1	9	6	1	26	1	1
Apo A1 (mg/dL)	96-176	143	188	177	152	135	135	94	136
Apo B (mg/dL)	43-128	61	93	125	78	60	72	<41	83

* Numbers and letters listed after proband/kindred number reflect the following: age (in years); sex (M = male, F = female), and race (B = black, W = white).

† Protein C data for kindred 2 displayed in Figure 1.

‡ Protein C deficiency.

§ Protein S deficiency.

|| ND, not done.

¶ LDL cholesterol <10th percentile (48).

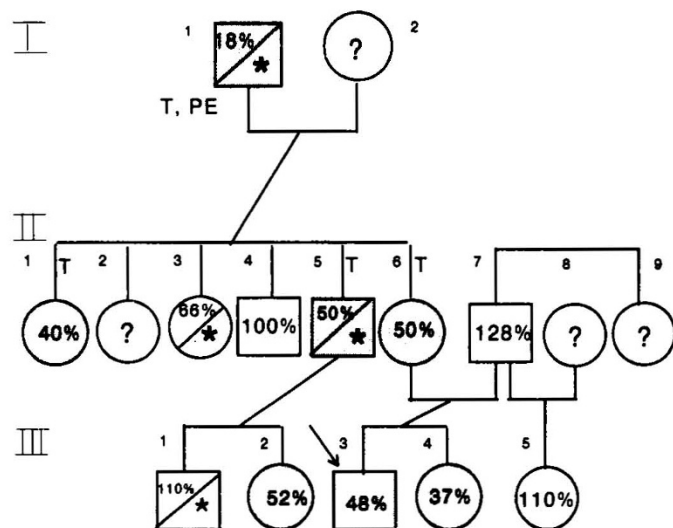


Fig. 1. Kindred 2. Three generations with familial protein C deficiency and with familial high PAI (10). The pediatric proband (→) with Legg-Perthes disease and protein C deficiency is III-3. Shaded symbols, Protein C deficiency; T, thrombosis or thrombophlebitis; PE, pulmonary embolus; *, high PAI-Fx and/or high PAI-Ag. Protein C levels are displayed as % of normal. Normal range, 55-145%.

decile) PAI-Fx, PAI-Ag, or both (Table 2, Fig. 1). High PAI (F_x, A_g, or both) and low tPA-Fx appeared to segregate independently of protein C deficiency (Table 2, Fig. 1). PAI-Ag and PAI-Fx, the major inhibitors of fibrinolysis (10, 11), were high in the proband's grandfather (I-1), aunt (II-3), uncle (II-5), and male first cousin (III-1) (Table 2, Fig. 1). Inability to activate fibrinolysis (10) (to normally increase tPA in response to venous occlusion) accompanied high PAI in the proband's aunt (II-3), in his male first cousin (III-1), and, unaccompanied by high PAI, was found in his female first cousin (III-2) (Table 2).

In kindred 6, where the proband was protein C deficient (Table 1), his 9-yr-old brother had normal protein C (78%), as did his 33-yr-old mother (69%). The proband's father was long separated from the family, and his whereabouts and health history were unknown. There was no history of thrombosis, thrombophlebitis, or Legg-Perthes disease in the proband's brother, mother, or maternal side of the family.

In kindred 7, where the proband was protein C deficient (Table 1), protein C levels were normal in the mother (93%), father (105%), and sister (100%). The mother and sister, like the proband, had familial hypobetalipoproteinemia with LDL cholesterol levels of 1.63 and 1.63 mmol/L respectively, and Stickler's syndrome. Beyond the proband's Legg-Perthes disease, there was no family history of thrombosis, thrombophlebitis, or osteonecrosis.

In kindred 8, beyond Legg-Perthes disease at age 6, the protein S-deficient proband had no history of thrombophlebitis or venous thrombosis despite major soft-tissue trauma from a recent severe automobile accident and from playing amateur soccer. In addition to the proband's very low levels of total protein S-Ag (16%; normal, 70-150%), and very low free protein S-Ag (<10%;

Table 2. Measurements of basal and stimulated fibrinolysis, lipids, and LP(a) in kindred 2 with familial protein C deficiency*

Kindred position	Age	Sex	Basal	Basal	Stimu-	Basal	Stimulated	TC	TG	HDL	LDL	Lp(a)
			PAI-Ag (ng/mL)	PAI-Fx (U/mL)	lated PAI-Fx (U/mL)	tPA-Fx (IU/mL)	tPA-Fx (IU/mL)					
Normal range			3.2–37.1	3.5–26.9	3.6–23.9	0.11–1.94	2.28–11.28	<5.17	<2.82	>0.91	<4.14	<20
Kindred position												
I-1	65	M	53.8†	11.0	10.9	0.38	3.16	6.10	2.01	1.27	3.90	53
I-2‡	68	F										
II-1	36	F	6.7	5.4	2.8	0.98	6.52	5.59	1.76	1.50	3.28	1
II-2‡	38	F										
II-3	42	F	109.6†	53.8†	63.0†	0.07§	2.16§	5.69	0.98	1.55	3.70	1
II-4	40	M	21.2	8.2	9.6	0.60	2.42	5.28	1.39	1.40	3.23	1
II-5	46	M	69.8†	47.7†	61.6†	0.23	3.46	8.46	1.46	1.86	5.92	1
II-6	33	F	23.0	10.0	10.2	0.51	3.03	4.55	.56	1.32	2.97	45
II-7	47	M	19.8	8.2	9.8	0.46	3.21	6.18	1.58	1.19	4.27	1
II-8‡	46	F										
II-9	49	F	24.9	17.0	14.4	0.42	4.77	5.74	2.64	1.14	3.39	64
III-1	21	M	53.3†	36.5†	40.7†	0.17	1.52§	6.00	1.31	1.58	3.83	17
III-2	19	F	10.8	5.8	8.4	0.50	1.04§	5.97	2.70	1.76	2.97	17
III-3	7	M	16.0	6.4	ND¶	2.48	ND	4.60	0.36	1.99	2.46	1
III-4	13	F	15.9	8.2	ND	0.60	ND	3.98	0.38	1.42	2.38	1
III-5	28	F	7.2	6.8	6.6	0.97	5.01	4.22	1.16	1.47	2.20	1

* Protein C levels (% of normal) are displayed in Figure 1.

† High (top decile) PAI.

‡ Unavailable for blood sampling.

§ Low (bottom decile) tPA-Fx.

|| Proband with Legg-Perthes disease.

¶ ND, not done.

normal, 65–135%), functional protein S was very low (<12.5%; normal, 62–138%), as was the C4B binding protein (73%; normal, 85–135%; Table 1). As may occur in familial protein S deficiency, the proband in kindred 8 failed to activate fibrinolysis (Table 1). His protein S-deficient brother had recurrent thromboses since age 19 y, and, despite long-term coumadin therapy, had sustained mesenteric vein thrombosis at age 43 y.

In proband 5, after 10 min of venous occlusion, no activation of fibrinolysis occurred despite normal PAI-Fx, PAI-Ag, Lp(a), and normal proteins C, S, and antithrombin III (Table 1). We do not yet understand why this proband could not initiate fibrinolysis. Family members were unavailable for study. The proband had no thrombotic events beyond Legg-Perthes disease. There was no family history of thrombosis, thrombophlebitis, or osteonecrosis.

All eight probands had normal basal PAI-Fx and normal basal α_2 -antiplasmin and fibrinogen (Table 1).

Basal plasminogen was inexplicably low in proband 3 (Table 1). Basal D-dimer levels were normal in the six probands where they were measured (Table 1).

Postocclusion blood could not be obtained in proband 2; postocclusion, activated tPA-Fx was normal in five of the remaining seven probands, being low in proband 5 and in proband 8, as above (Table 1).

DISCUSSION

In adults, we have recently shown that very high PAI with inability to activate fibrinolysis is a common cause of idiopathic osteonecrosis, whereas high Lp(a) or protein C deficiency may play an etiologic role in secondary osteonecrosis (10, 11). When we turned our attention to Legg-Perthes disease (1–9, 18–21, 34), we postulated that we might again find high PAI with consequent inability to activate fibrinolysis (10, 11). However, in the current study, only one child could not initiate fibrinolysis, and he unexpectedly had normal PAI-Ag, PAI-Fx, and Lp(a), as well as

normal proteins C, S, and antithrombin III. We do not yet know why he was unable to initiate fibrinolysis.

Of the eight patients with Legg-Perthes disease, four (50%) had thrombophilic disorders (12–17, 27, 28, 38–44), three had primary thrombophilia with previously undiagnosed deficiency of protein C, and one had familial protein S deficiency. Protein C and protein S deficiency, transmitted as autosomal dominant disorders (38, 40), are rare both in free-living, unselected population groups (12–14) and among cohorts selected because of thrombosis or thrombophlebitis, where they account for only a small percentage (usually <5% altogether) of cases (17, 38–44). Hence, our finding that 50% of probands with Legg-Perthes *also* had protein C or S deficiency is exceptionally higher than would be suggested by chance (1 in 325 000) and implies causality, particularly because our probands with Perthes were sampled solely on the basis of their orthopedic diagnoses, their current residence in the Cincinnati area, and their willingness to participate in the study.

Given the remarkably low estimated likelihood (1 in 325 000) that unselected children would have protein C or S deficiency and concurrent Legg-Perthes disease, a prospective study of children without symptoms of heterozygous for protein C deficiency to determine how many would have Legg-Perthes disease develop would be expensive and impractical. It would first require population screening of healthy children to identify those [1 in 250 (14) to 1 in 16 000 (13)] with protein C deficiency, with subsequent follow-up through the end of skeletal maturation.

In one of the three kindreds where the proband had protein C deficiency, there was three-generation vertical and horizontal transmission of low protein C accompanied by thrombophlebitis and pulmonary emboli in protein C-deficient adults, characteristic of autosomal dominantly transmitted protein C deficiency (16, 38–44). All four protein C-deficient adults had thrombotic events but not osteonecrosis. Legg-Perthes disease apparently represented the thrombotic event in the pediatric proband. Unexpectedly, this kindred also had three-generation vertical and

horizontal transmission of high PAI-Fx and/or high PAI-Ag, another dominantly transmitted trait that causes hypofibrinolysis and idiopathic osteonecrosis in adults (10, 11). The high PAI appeared to segregate independently of protein C deficiency. We have not found any reports of familial protein C-deficient kindreds who also had concurrent familial hypofibrinolysis (mediated by high PAI) (10, 11).

In two kindreds identified by a protein C-deficient pediatric proband, familial protein C deficiency could not be demonstrated. In one of these two kindreds, the proband's father, the likely protein C-deficient carrier, was unavailable. In the second kindred, both parents and a sibling unexpectedly (14-16) had normal protein C.

In the kindred with protein S deficiency, the proband's S-deficient brother had sustained mesenteric venous thrombosis and, characteristically (17, 27, 28, 38, 40), began to have thrombotic events at age 19 y.

Presumably, thrombophilia associated with deficient protein C and S in childhood, in a fashion similar to hypofibrinolysis mediated by high PAI and/or by high Lp(a) in adults (10, 11), leads to venous thrombi in the femur with subsequent obstruction of bone venous drainage, venous hypertension, osteonecrosis, and Legg-Perthes disease (8, 9). We do not know what factors in protein C- and S-deficient families cause some affected family members to have Legg-Perthes disease and some to have other sites for thrombosis (thrombophlebitis, for example).

The finding of protein C or S deficiency as common pathophysiologic causes of Legg-Perthes disease should open avenues to family diagnosis and to therapy. Children with Legg-Perthes syndrome should be systematically evaluated for protein C and S deficiency and hypofibrinolysis. If thrombophilic, hypofibrinolytic disorders are found, all family members should be screened (16, 38).

In one kindred, protein C deficiency and familial hypobetalipoproteinemia (35) were both present in the proband. It is possible that the gene defects for apo B [responsible for familial hypobetalipoproteinemia (45-47)] and protein C (45) are linked on chromosome 2, an area deserving of further study. Protein C deficiency is transmitted as an autosomal dominant trait, with the gene located on chromosome 2 (q13-14) (45). Familial hypobetalipoproteinemia is transmitted as an autosomal dominant trait, with the apo B gene also located on chromosome 2 (45-47). No published data exist suggesting that low total or LDL cholesterol could affect protein C levels or vice versa, and we postulate that the association of protein C deficiency and familial hypobetalipoproteinemia is genetic.

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