

Bianca Bassi
Emilia Parodi
Maria Messina
Patrizia Boffi
Barbara Bobba
Maria Francesca Campagnoli
Roberto Rigardetto
Paola Saracco

Screening for genetic and acquired thrombophilia in a cohort of young migrainous patients

Received: 31 October 2002
Accepted in revised form: 17 February 2003

B. Bassi • P. Boffi • B. Bobba • R. Rigardetto
Division of Child Neuropsychiatry,
University of Turin,
Regina Margherita Hospital, Turin, Italy

E. Parodi • M.F. Campagnoli • P. Saracco (✉)
Pediatric Hematology Unit,
University of Turin,
Regina Margherita Hospital,
Piazza Polonia 94, I-10126 Turin, Italy
e-mail: saracco@pediatria.unito.it
Tel.: +39-011-3135259
Fax: +39-011-3135382

M. Messina
Immunohematology and Transfusion Medicine,
University of Turin,
Regina Margherita Hospital, Turin, Italy

Abstract Growing evidence suggests a possible relationship between migraine and thrombotic risk factors. The aim of this study was to analyze the possible relationship between migraine and acquired and genetic thrombophilia in a young population. We compared 16 migrainous adolescents, 12 children with tension-type headache, and controls in terms of frequencies of prothrombotic polymorphisms (factor V Leiden, C677T mutation of 5,10 methyl-enedetetrahydrofolate reductase, G20210A mutation of prothrombin), platelet aggregability, anticoagulant antibodies, blood lipid pattern, serum folate and vitamin B12

levels, homocysteinemia, coagulation parameters, and family history for migraine and precocious thrombotic events. This study confirms the link between migraine and increased platelet responsiveness. Overall, 62.5% of migrainous patients carried at least three thrombophilic factors. Our preliminary data suggest that, in order to assess prevention strategies, it could be appropriate to perform a complete thrombophilia screening in young patients suffering from migraine and with a family history of thrombosis.

Key words Thrombophilia • Migraine • Childhood

Introduction

Primary headache is a common complaint in pediatric medicine. Bille [1] demonstrated that 40% of children have significant headache episodes before age 7 years and 75% before age 15 years. The prevalence of migraine is 1.4% and 5.3% before age 7 and age 15 years, respectively [2]. During childhood there is no significant difference between the sexes; after puberty migraine is more common in girls.

A positive family history is a well established risk factor for migraine, even if the modality of genetic transmission is still unrecognized. Migraine is a polygenic multifactorial disease, as both environmental and genetic factors, with

multiple genes interacting, are involved in its pathogenesis [3]. Neurogenic inflammation has been proposed as a possible mechanism for migraine, with proinflammatory mediators such as interleukin-1-alpha playing an important role [4]. Platelet activation leading to, among other things, the release of thromboxane, has also been implicated [5, 6]. During migraine attacks, both increased platelet aggregability [7] and plasma coagulability [8] have been described, and growing evidence suggests a possible relationship between migraine and both inherited and acquired thrombotic risk factors.

Epidemiological studies have shown a consistent association between migraine and ischemic stroke [9–14]. Migraine has also been considered to be an independent risk

factor for stroke in the young [15], with implications for patient management.

Within the past decade, various mutations in genes encoding proteins regulating blood coagulation have been shown to play a major role in predisposition to thrombosis. Most of these genetic variations are risk factors for venous thrombosis and have minor role in arterial thrombosis: protein C, protein S and antithrombin deficiencies, activated protein C (APC) resistance mainly due to factor V Leiden mutation, and factor II G20210A mutation [16]. More recently, other inherited defects have been suspected to play a role in arterial thrombosis: platelet receptor polymorphisms (C807T allele of GPIa/IIa, HPLA1 and HPLA2 of GPIIIa, GPIb alpha), hyperhomocysteinemia due to mutation C677T of 5, 10-methylenetetrahydrofolate reductase (MTHFR) gene, and cystathionine β synthetase deficiency, as well as high levels of factors VII and VIII. Other genetic defects are under investigation as risk factors: mutation G455A of the β chain of fibrinogen, polymorphism of ApoE, polymorphism of ACE gene, polymorphism 4G/5G of PAI-1 gene, polymorphism of thrombomodulin, polymorphism A353G of factor VII, and mutation of TFPI gene [17].

Among acquired conditions, antiphospholipid antibodies are the major risk factor for both venous and arterial thromboses.

Few studies have investigated the prevalence of prothrombotic genetic risk factors in patients with migraine, and conflicting results have been reported. A Finish study detected a higher frequency of migraine with aura in patients suffering from ischemic stroke who carried the factor V Leiden (67%) than in those who had no mutation (26%) [18]; these findings were subsequently confirmed by Leone et al. [19]. In contrast, a case-control study did not find a significantly higher frequency of several prothrombotic factors (factor V Leiden, G20210A mutation of factor II, decanucleotide insertion/deletion in factor VII promoter, HPA-1 and HPA-2 polymorphisms) in 106 migrainous patients compared to healthy controls [20], even if a particularly high frequency of factor V Leiden, not reaching statistical significance, was found in patients suffering from migraine with aura. D'Amico et al. [21] compared the frequency of genetic abnormalities of protein C system in three groups: patients with migraine with aura, patients with ischemic stroke, and healthy controls. They found a significantly higher frequency of activated protein C (APC) resistance due to factor V Leiden and protein S deficiency in migraine and ischemic stroke patients than in controls. These findings were not confirmed by Soriani et al. [22] who investigated the frequency of factor V Leiden in a cohort of young migrainous patients aged 8–28 years. More recently, a higher frequency of C677T mutation of MTHFR in migraine versus healthy controls has been detected by Kowa et al. [23].

Thus, pathophysiological studies aimed at identifying the role played by various prothrombotic factors in the ischemic cascade are recommended. In the present study we

assessed the prevalence of several prothrombotic risk factors, both inherited and acquired, in a population of migrainous children, mainly adolescents.

Subjects and methods

The study enrolled all children complaining of headache episodes admitted consecutively to the Center for Diagnosis and Care of Headache of the Department of Child Neuropsychiatry (University of Turin) between September 2000 and September 2001. Diagnosis of migraine and tension-type headache was made according to the criteria of the International Headache Society [24].

A group of age-matched children awaiting minor surgery (excluding adenotonsillectomy and appendectomy) served as controls for most procedures, except for measuring the frequency of the three prothrombotic polymorphisms. For this test, the control group consisted of 145 children with malignant disease who were screened for thrombophilia before insertion of a central venous line to chemotherapy.

Informed consent was obtained from the parents of all the children.

The parents of every patient were carefully asked about family history for migraine and about the onset of thrombosis or cerebrovascular disease before age 50 years in first-degree relatives.

Biochemical and genetic analyses

Venous blood from headache children was withdrawn during headache-free period (at least 5 days from an attack) and not in concomitance with drug intake.

The following analyses of prothrombotic factors were performed in all subjects: coagulation tests including physiologic anticoagulants, anticoagulant antibodies, metabolic tests (blood lipid pattern, serum folate and vitamin B12 levels, basal homocysteinemia), platelet function assay and assay of prothrombotic polymorphisms for factor V Leiden, factor II and MTHFR. During childhood, the development of a thromboembolic event is usually dependent on the simultaneous presence of at least three different prothrombotic factors [25], both inherited and acquired. Thus, a transverse analysis was performed in order to evaluate the frequency of three or more risk factors in all patients.

The following tests were performed in all subjects (methods in parentheses): prothrombin time (PT) and international normalized ratio (INR), activated partial thromboplastin time (APTT) and APTT ratio, fibrinogen (*Clauss method*), D-dimer (*turbidometric determination*), antithrombin III (*chromogenic determination*), protein C (*chromogenic determination*), protein S functional activity, APC resistance (*protein C activity dependent clotting time*), factors VII, VIII:c, XI, XII, VIII Von Willebrand (*ristocetin cofactor activity*) and plasminogen (*chromogenic determination*).

The IgG and IgM forms of antiphospholipid antibodies (APA) and anticardiolipin antibodies (ACA) were also assayed.

Furthermore, plasma levels of triglycerides and total and HDL cholesterol were assessed by enzymatic method. Apolipoproteins A and B were assayed by immunoturbidimetric method. Plasma levels of homocysteine were detected by HPLC. Serum concentrations of folate and vitamin B12 were analyzed by radioimmunoassay.

Platelet aggregation, induced exogenously (ADP, 1 and 3 μ M; epinephrine, 0.5 and 20 μ M; collagen 2 μ g/ml; ristocetin 1.5 μ g/ml; arachidonic acid, 0.5 μ M), was determined according to the optical aggregometer method of Born

DNA was extracted from peripheral blood mononuclear cells by standard methods. The G to A transition at nucleotide 1691 of the factor V gene (factor V Leiden), the G to A transition at nucleotide 20210 of the prothrombin gene (factor II) and the C677T substitution of the 5, 10-methylenetetrahydrofolate reductase (MTHFR) gene were analyzed by polymerase chain reaction (PCR) amplification, digestion by restriction enzymes (respectively, Mnl I, Hinf I and Hind III) and agarose gel electrophoresis.

Transverse analysis of risk factors

We considered as a single prothrombotic factor any of the following:

- A positive family history of precocious thrombotic events;
- Any prothrombotic polymorphism (heterozygous or homozygous);
- Basal hyperhomocysteinaemia (more than 20 μ mol/l)
- Deficiency of any physiologic anticoagulant (ATIII less than 70%, protein C less than 50%, or protein S less than 60%)
- APC resistance less than 0.86 (ratio)
- Factor VII over 130%, factor VIII over 150%, or fibrinogen over 400 mg/dl
- Platelet hyperaggregation (Born test) to at least one agonist (ADP, epinephrine, collagen, ristocetin, arachidonic acid)
- Presence of anticoagulant antibodies

Statistical analysis

Differences between groups were assessed using Student's *t* test, when appropriate. The prevalence of prothrombotic genetic risk factors in the different groups was compared by chi-square analysis. A value of $p < 0.05$ was considered to be significant.

Results

The study enrolled 28 children with headache (migraine or tension-type) and 28 age-matched controls. The migraine group comprised 16 patients, including 8 boys and 8 girls, aged 11–17 years; mean age of the boys was 13.1 years, while that of the girls was 13.6 years. Nine children had migraine

with aura and 7 had migraine without aura. None had basilar or familiar hemiplegic migraine. Four of 8 boys and 2 of 8 girls were prepubertal. The tension type headache group consisted of 12 children (7 boys and 5 girls) aged 8–13 years; mean age of the boys was 11.7 years while that of the girls was 10 years. Among the 7 boys, 3 were prepubertal while 3 of 5 girls were prepubertal. No girl was taking estroprogestational drugs. Only two adolescents were occasional smokers.

The extent of platelet aggregation induced by ADP (at low concentration) and epinephrine (at low and high concentrations) was significantly higher in samples from migrainous patients than from controls ($p < 0.05$). Additionally, a significant difference in epinephrine-induced aggregation (at both concentrations) was observed between samples from migrainous children and tension-type headache subjects ($p < 0.05$) (Fig. 1). Within the migrainous population, age (less or more than 12 years), puberty, gender or the presence of aura were not related with higher platelet aggregation (data not shown).

One migrainous child, with family history for thrombosis, fulfilled diagnostic criteria for sticky platelet syndrome, having hyperaggregation induced by low concentrations of both ADP and epinephrine.

The extents of platelet aggregation induced by other agonists were comparable among the two patient groups and controls (data not shown).

Coagulation parameters, antibodies and metabolic tests measured in the groups of migrainous and tension-type headache children are given in Table 1. All values fell within normal range. Values from the control group were not significantly different from those of the two patient groups (data not shown).

One child suffering from migraine and one with tension-type headache presented a mild (50%) deficiency of protein S; none of the controls had any deficit of physiologic anticoagulants.

The factor V Leiden mutation was not found in any of the patients with migraine, while it was found in heterozygous form in 2 (16%) of 12 patients with tension-type headache and in 2 (1.4%) of 145 controls ($p > 0.05$) (Table 2). The factor II G20210A allele was observed in 1 migraineur (6.2%), in no children with tension-type headache and in 4 controls ($p > 0.05$). The polymorphism C677T of MTHFR was detected in homozygous form in 5 patients (31%) with migraine, in 3 children (25%) suffering from tension-type headache and in 16 controls (11%). The difference between migraine group and controls was significant ($p < 0.05$).

Transverse analysis showed that all 28 children investigated had at least one prothrombotic risk factor (Table 3). Among the 16 migrainous patients, 8 (50%) carried 3 or more thrombophilic factors. This percentage rose to 62.5% (10 of 16) when we also considered the presence of aura as risk factor. Of the 12 children with tension-type headache (Table 4), 5 (41%) presented at least 3 altered parameters ($p > 0.05$).

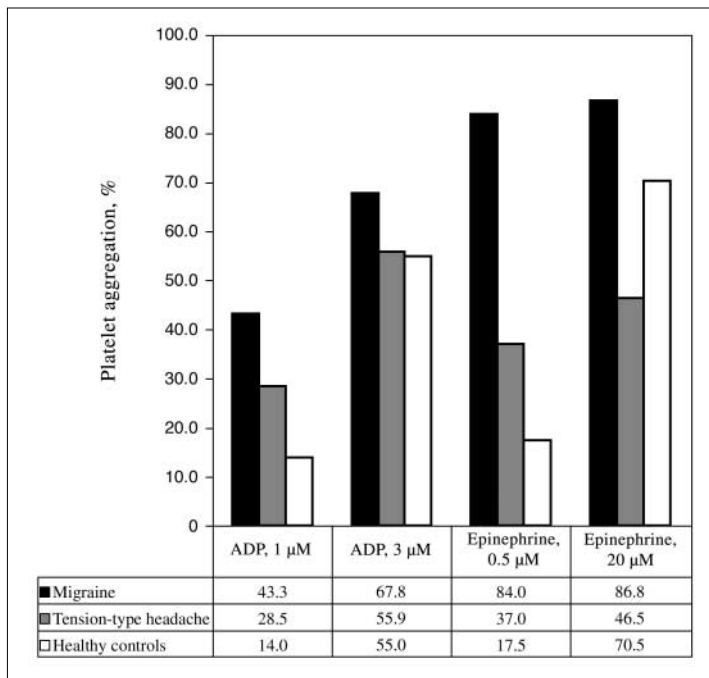


Fig. 1 Platelet aggregation induced by ADP and epinephrine in samples from migrainous and tension-type headache patients and from controls

Table 1 Coagulation and metabolic parameters in patients with migraine and tension-type headache. Values are mean (SD)

Test	Normal range	Migraine	Tension-type
INR	0.86-1.27	1.10 (0.16)	1.02 (0.08)
Fibrinogen, mg/dl	200-400	273 (89)	300 (65)
D-Dimer, μg/ml	0-0.25	0.24 (0.16)	0.21 (0.10)
AT III, %	70-120	104 (11)	105 (15)
Protein C, %	50-140	103.3 (19.2)	110.8 (23.8)
Protein S, %	60-130	89.47 (19.64)	91.73 (18.49)
APC resistance ratio	0.86-1.2	1.04 (0.08)	0.87 (0.16)
Factor VII, %	70-130	92.6 (19.6)	104 (29)
Factor VIII:c, %	60-150	121.4 (40.5)	126.0 (35.4)
Factor VIII, %	58-166	108.5 (59.0)	137.2 (67.3)
Factor XI, %	60-140	94.25 (16.32)	99.30 (23.45)
Factor XII, %	70-150	92.57 (25.82)	97.27 (30.87)
Plasminogen, %	70-150	93.33 (13.33)	106.18 (22.45)
APA Ig G, UI/ml	0-10	1.61 (2.45)	2.01 (3.49)
APA IgM, UI/ml	0-3	1.91 (3.64)	0.64 (0.97)
ACA IgG, UI/ml	0-5	3.63 (2.52)	3.72 (4.09)
ACA IgM, UI/ml	0-5	3.41 (2.16)	2.87 (4.33)
Triglycerides, mg/dl	<150	58.57 (18.67)	66.2 (34.92)
Total cholesterol, mg/dl	<200	162.4 (25.4)	173.3 (21.4)
HDL cholesterol, mg/dl	>45	57.93 (11.2)	56.1 (15.6)
Apolipoprotein A, mg/dl	105-205	140.5 (21.0)	143.2 (18.8)
Apolipoprotein B, mg/dl	55-130	69.3 (0.09)	72.54 (14.46)
Homocysteine, μM	6.78-20.00	6.25 (1.13)	5.97 (1.83)
Folate, ng/ml	2-16	6.14 (3.03)	6.16 (2.95)
Vitamin B12, pg/ml	200-800	510.84 (153.49)	503.77 (174.59)

APA, antiphospholipid antibodies; ACA, anticardiolipin antibodies; INR, international normalized ratio; AT III, antithrombin III

Table 2 Frequency of 3 polymorphisms (factor V Leiden heterozygous, C677T of MTHFR homozygous, G20210A of prothrombin heterozygous) in migraine and tension-type headache children and in controls. Values are n (%) of subjects

	Factor V Leiden (heterozygous)	C677T MTHFR (homozygous)	G20210A prothrombin (heterozygous)
Migraine, n=16	0 (0)	5 (31)	1 (6.2)
Tension-type headache, n=12	2 (16)	3 (25)	0 (0)
Controls, n=145	2 (1.4)	16 (11)	4 (2.7)

All patients were reassessed six months later for clinical and laboratory follow-up, in order to check the previously detected abnormal parameters. The increased platelet responsiveness was confirmed in all patients with a previously altered Born test. The clinical follow-up showed that one boy suffering from migraine with aura subsequently had a transitory ischemic attack; he presented three risk factors: homozygous MTHFR, platelet hyperaggregation to low and high ADP and epinephrine, and a family history for arterial thrombosis.

A total of 10 migrainous patients (63%) and 5 with tension-type headache (42%) presented a family history for migraine. Furthermore, parents of 4 migraineurs (25%) and 3 children with tension-type headache (25%) referred pre-

mature onset of thrombosis and cerebrovascular disease in first-degree relatives.

Discussion

The aim of this study was to analyze the possible relationship between migraine and acquired and genetic prothrombotic factors in a young population, in which the influence of other behavioral and age-dependent risk factors (such as smoking habit, alcohol consumption and contraceptive drug use) was excluded. The study confirms the association between migraine and increased platelet responsiveness.

Table 3 Presence of prothrombotic risk factors in 16 children with migraine

	Patient																Total, n (%)
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	
Migraine with aura		x	x	x	x	x	x		x				x	x			9 (56)
Family history of thrombosis	x					x					x			x			4 (25)
G20210A factor II polymorphism						x											1 (6)
C677T MTHFR polymorphism		x	x	x		x	x		x	x	x	x					9 (56)
Low protein S			x														1 (6)
High factor VII						x				x						x	3 (18)
High factor VIII				x				x		x				x	x		5 (31)
High fibrinogen			x						x		x					x	4(25)
Platelet hyper-aggregation		x			x	x		x	x			x		x			7 (44)
Antiphospholipid or anticardiolipin antibodies			x	x								x		x	x	x	6(37)
Total number of risk factors for each patient	1	3	5	4	2	6	2	2	4	3	3	3	1	5	4	1	

Table 4 Presence of prothrombotic risk factors in 12 children with tension-type headache

	Patient												Total, n (%)
	1	2	3	4	5	6	7	8	9	10	11	12	
Family history of thrombosis			x		x						x		3 (25)
Leiden polymorphism			x			x							2 (16)
C677T MTHFR polymorphism	x	x	x	x	x		x	x	x	x	x		10 (83)
Low protein S	x												1 (8)
APC resistance						x					x		2 (16)
High factor VII								x					1 (8)
High factor VIII				x				x			x		3 (25)
High fibrinogen					x								1 (8)
Platelet hyper-aggregation		x			x								2 (16)
Antiphospholipid or anticardiolipin antibodies						x	x					x	3 (25)
Total number of risk factors for each patient	2	2	3	2	4	3	2	3	1	1	4	1	

Although responses varied depending on the aggregation agent used, the data agree with previous studies showing abnormal platelet behavior also during the headache-free period. On the other hand, in contrast with Tozzi et al. [26] and Kovacs et al. [7], who found a common trend towards platelet hyperaggregation in both tension-type headache sufferers and in migrainous patients, we detected a significant difference between the two groups. We concluded that platelet hyperaggregation may be considered to be a more specific marker of migraine.

Conflicting results have been reported regarding the prevalence of genetic prothrombotic risk factors in migraine [18–23]. In our study, in accordance with Corral et al. [20] and Soriani et al. [22], the frequencies of the two main prothrombotic polymorphisms for venous thromboembolism (Leiden, prothrombin) did not differ from those of controls. Homozygous MTHFR thermolabile variant was detected at a significantly higher frequency in the group of migrainous children than in controls, in accordance with Kowa et al. [23].

We also did not find any difference in anticoagulant antibody levels in migrainous children compared with control subjects. These data agree with previously studies in children [27, 28] and, recently, in adults [29]. The role of these

antibodies in migraine remains to be elucidated. Robbins [30] found anticardiolipin IgG in 22 of 68 migrainous patients and only in 1 of 22 healthy controls. A significant association between anticoagulant antibodies and stroke during childhood has been reported [31, 32].

Our transversal analysis showed that a high percentage of children with migraine and tension-type headache carried at least three risk factors for thrombosis, including a positive family history. However, considering the presence of aura as an additional risk factor, this percentage rose to 62.5% in the migraine group. These data, together with the increased platelet responsiveness, suggest a possible higher risk of developing thrombotic events in these children. For this reason, prophylactic treatment with platelet anti-aggregants (such as low dose aspirin) could be considered in patients with persistent hyperaggregability and other prothrombotic factors, including positive family history. In ongoing prospective study, we are further analyzing arterial prothrombotic genetic risk factors (such as platelet protein polymorphisms) and other platelet activation markers (such as p-selectin and alpha1/beta1 protein expression) in selected young patients with migraine. This study may give us useful information on the role of platelet behavior and arterial hemostasis in migraine.

In conclusion, despite the limited size of the study, our preliminary data suggest that it could be appropriate to perform a complete thrombophilia screening in young patients suffering from headache (particularly migraine with aura) and/or with a family history of thrombosis.

Additional randomized studies are warranted to evaluate the efficacy of prophylactic treatment with platelet anti-aggregants in migrainous patients considered to be at risk for thromboembolic events or having persistent platelet activation.

References

- Bille B (1962) Migraine in school-children. *Acta Paediatr* 51(136):1–151
- Linder SL, Winner P (2001) Pediatric headache. *Med Clin North Am* 85:1037–1053
- Montagna P (2000) Molecular genetics of migraine headaches: a review. *Cephalalgia* 20:3–14
- Rainero I, Pinessi L, Salani G, Valfre W, Rivoiro C, Savi L, Gentile S, Giudice RL, Grimaldi LM (2002) A polymorphism in the interleukin-1 alpha gene influences the clinical features of migraine. *Headache* 42(5):337–340
- Kitano A, Shimomura T, Takeshima T, Takahashi K (1994) Increased 11-dehydrothromboxane B2 in migraine: platelet hyperfunction in patients with migraine during headache-free period. *Headache* 34(9):515–518
- Puig-Paralada P, Planas JM, Gimenez J, Sanchez J, Gaya J, Tolosa E, Obach J (1991) Plasma and saliva levels of PGI2 and TXA2 in the headache-free period classical migraine patients. The effects of nicardipine. *Headache* 31(3):156–158
- Kovacs H, Herman F, Filep J, Jelencsik I, Magyar K, Csanda E (1990) Platelet aggregation of migraineurs during and between attacks. *Cephalalgia* 10:161–165
- Kalendovsky Z, Austin JH (1975) “Complicated migraine” its association with increased platelet aggregability and abnormal plasma coagulation factors. *Headache* 15(1):18–35
- Tzourio C, Tehindrazanarivelo A, Iglesias S, Alperovitch A, Chedru F, d’Anglejan-Chatillon J, Bousser MG (1995) Case-control study of migraine and risk of ischaemic stroke in young women. *BMJ* 310:830–833
- Carolei A, Marini C, De Matteis G and the Italian National Research Council Study on Stroke in the Young (1996) History of migraine and risk of cerebral ischaemia in young adults. *Lancet* 347:1503–1506
- Merikangas KR, Fenton BT, Cheng SH, Stolar MJ, Risch N (1997) Association between migraine and stroke in a large scale epidemiological study of the United States. *Arch Neurol* 54:362–368
- Chang CL, Donaghy M, Poulter N (1999) Migraine and stroke in young women: case-control study. *BMJ* 318:13–18
- Tzourio C, Kittner SJ, Bousser MG, Alperovitch A (2000) Migraine and stroke in young women. *Cephalalgia* 20:190–199
- Parnetti L, Paciaroni M, Gallai V (2002) Headache and ischemic stroke. *J Headache Pain* 3:15–20
- Martin PJ, Enevoldson TP, Hymphrey PR (1997) Causes of ischaemic stroke in the young. *Postgrad Med J* 73(855):8–16
- Selighson U, Lubetsky A (2001) Genetic susceptibility to venous thrombosis. *N Engl J Med* 344(16):1222–1231
- Simmonds RE, Hermida J, Rezende SM, Lane DA (2001) Haemostatic genetic risk factors in arterial thrombosis. *Thromb Haemost* 86:374–805
- Kontula K, Ylikorkala A, Miettinen H, Vuorio A, Kauppinen-Makelin R, Hamalainen L, Palomaki H, Kaste M (1995) Arg506Gln factor V mutation (factor V Leiden) in patients with ischaemic cerebrovascular disease and survivors of myocardial infarction. *Thromb Haemost* 73:558–580
- Leone M, Ciusani E, Ariano C, Moschiano F, Grazi L, D’Amico D, Nespolo A, Bussone G (1996) Point mutation (Arg506Gln) of factor V in migraine with and without aura. *Cephalalgia* 16:395 (abstract)
- Corral J, Iniesta JA, Gonzales-Conejero R, Lozano ML, Riviera J, Vicente V (1998) Migraine and prothrombotic genetic risk factors. *Cephalalgia* 18(8):257–260
- D’Amico D, Moschiano F, Leone M, Ariano C, Ciusani E, Erba N, Grazi L, Ferraris A, Schieroni F, Bussone G (1998) Genetic abnormalities of the protein C system: shared risk factors in young adults with migraine with aura and with ischemic stroke? *Cephalalgia* 18:618–621
- Soriani S, Borgna-Pignatti C, Trabetti E, Casartelli A, Montagna P, Pignatti PF (1998) Frequency of factor V Leiden in juvenile migraine with aura. *Headache* 38:779–781
- Kowa H, Takeshima T, Urakami K, Sakai F, Nakashima K (2000) The homozygous C677T mutation in the methyltetrahydrofolate reductase gene is a genetic risk factor for migraine. *Am J Med Genet* 96(6):762–764
- (1988) Classification and diagnostic criteria for headache disorders, cranial neuralgias and facial pain. Headache Classification Committee of the International Headache Society *Cephalalgia* 8[Suppl. 7]:1–96
- Andrew M (2000) Congenital prothrombotic disorders: presentation during infancy and childhood. In: Andrew M, Monagle PT, Brooker L (eds) *Thromboembolic complications during infancy and childhood*. B.C. Decker, Hamilton London, pp 47–110

-
26. Tozzi E, Tozzi-Ciancarelli MG, Di Massimo C, Mascioli A, Angelini R, de Matteis F (1996) Platelet responsiveness in migrainous children during headache-free period. *Headache* 36:95–99
 27. Lanzi G, Zambrino CA, Ferrari-Ginevra O et al (1999) Anticorpi antifosfolipidi ed emicrania in età evolutiva. Atti del XIV Congresso Nazionale della SISC, Perugia, 19–22 September 1999 (abstract)
 28. Verrotti E, Cieri F, Pelliccia P Morgese G, Chiarelli F (2000) Lack of association between antiphospholipid antibodies and migraine in children. *Int J Clin Lab Res* 30:109–111
 29. Tietjen GE, Day M, Norris L, Aurora S, Halvorsen A, Schultz LR, Levine SR (1998) Role of anticardiolipin antibodies in young persons with migraine and transient focal neurologic events. *Neurology* 50:1433–1440
 30. Robbins L (1991) Migraine and anticardiolipin antibodies—case reports of 13 patients and the prevalence of antiphospholipid antibodies in migraineurs. *Headache* 31:357–359
 31. Olson JC, Konkol RJ, Gilli JC, Dobyms WB, Coull BM (1994) Childhood stroke and lupus anticoagulant. *Pediatr Neurol* 10:54–57
 32. Angelini L, Ravelli A, Caporali R, Martini A (1994) High prevalence of antiphospholipid antibodies in children with idiopathic cerebral ischemia. *Pediatrics* 87:933–935