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Features of essential thrombocythaemia in childhood: a study of five children

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Summary. Essential thrombocythaemia (ET) is usually considered a disease of the middle-aged but, with the advent of automated platelet counting, ET is diagnosed with increasing frequency in young adults and, even more rarely, in children. We report five paediatric patients (four girls and one boy, mean age 89 months) diagnosed with ET in agreement with Polycythaemia Vera Study Group criteria. The patients had a persistent thrombocytosis over 900×10^9/l and, at the time of diagnosis, their platelet count ranged between 1112 and 3178×10^9/l. A 9-month-old girl had thrombosis of the inferior cava vein, two children had headaches and two others remained asymptomatic throughout the follow-up period. Megakaryocytes in the bone marrow were increased in number. The chromosomal analysis was normal, and bcr rearrangement was always negative. None of the patients had spontaneous BFU-E or altered levels of serum erythropoietin and thrombopoietin. Two patients showed alteration of platelet aggregation, and all had decreased levels of intraplatelet serotonin. In spite of the diagnosis of ET in our patients, we are not sure that the cases reported here are really myeloproliferative disorders. The features could suggest that the cases observed may be affected by an 'idiopathic thrombocytosis' without myeloproliferation. Possible variants of ET are described in young adults, and the heterogeneous nature of ET is also suggested by our paediatric patients. Only careful long-term follow-up of patients such as these will clarify the natural history of these disorders and suggest therapeutic management.

Keywords: essential thrombocythaemia, thrombocytosis, childhood, chronic myeloproliferative disorders.

Essential thrombocythaemia (ET) is a chronic myeloproliferative disorder (MPD) variant (Kutti & Wadenvik, 1996). ET has usually been considered a disease of the middle-aged, having a slight preponderance in women, with an onset in the fifth or sixth decade of life (Van Genderen & Michiels, 1993). However, with the advent of automated platelet counting, increased platelet counts and, in particular, ET are diagnosed with increasing frequency in young adults and even in childhood, although less commonly than in adults. The 11 cases of ET in children reported in the literature (Michiels & Van Genderen, 1997) showed persistently elevated platelet counts over 1000×10^9/l with thrombotic and haemorrhagic episodes occurring in eight out of 11 patients.

We report our experience of five cases of ET in childhood, diagnosed in our department and followed over a 16-year period.

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PATIENTS AND METHODS

Five paediatric patients, observed in our department over the last 16 years, were diagnosed with ET in agreement with Polycythaemia Vera Study Group (PVSG) criteria (Murphy, 1983). There were four girls and one boy, aged 9–129 months, with a mean age of 89 months at the time of diagnosis. A persistent thrombocytosis over 900×10^9/l was repeatedly documented without any demonstrable cause of reactive thrombocytosis. In particular, infectious disease, asplenia, sideropenia or tumours were not demonstrated. Ultrasonography ruled out asplenia, and measurement of serum iron (examination of plasma iron, transferrin and ferritin) was used to assess the iron status.

Complete haematological evaluation of peripheral blood and bone marrow (morphological and cytochemical studies of peripheral blood and bone marrow and histological evaluation of trephine bone biopsy) was performed in all patients.

Karyotypes were evaluated by classical cytogenetic analysis on bone marrow samples (Sainati *et al*, 1997). All

our patients were analysed by polymerase chain reaction (PCR) for the presence of bcr/abl fusion transcript (primers: BCR-blA; BCR-e1A2; ABL-a3B; Schrappe *et al*, 1998). In one case (case D) M-bcr/abl gene fusion was also searched for by fluorescence *in situ* hybridization (FISH; Sainati *et al*, 1999).

In all these patients, spontaneous erythroid colony (BFU-E) was investigated using Iscove's method (Iscove *et al*, 1974). Serum erythropoietin level was evaluated by means of a commercial kit (EPO ELISA; Boehringer Mannheim, Biochemia, Milan, Italy) and expressed in U/l. Serum thrombopoietin (TPO) was evaluated in four out of the five patients with a commercial kit (Quantikine, ELISA, RD system Europe, UK; normal values 35 ± 22 pg/ml). Platelet aggregation with 2 μmol/l ADP, 2 μmol/l adrenaline and 2 mg collagen was performed according to Born's method (Fabris *et al*, 1984). Platelet serotonin content (5-HT) was performed as described previously (Fabris *et al*, 1984) by a fluorimetric method.

RESULTS

The main clinical and laboratory characteristics of our patients are summarized in Table I. In three patients, thrombocytosis was recognized incidentally. One patient was discovered because of headache and one, under evaluation for failure to thrive, was found to have thrombosis of the inferior cava vein.

Aspirin (ASA 100 mg/day) was used to treat patients with headache and was stopped if bleeds occurred (Randi *et al*, 1999a). Patient D was treated over a period of 2·5 years with alpha-interferon (IFN; median dose 2×10^6 U/three times a week), as prescribed by another physician.

In all patients, platelet counts remained high for a long time after initial observation. The median follow-up was 103·4 months (range 36–195 months). Continuous spontaneous remission was never achieved.

The parents and grandparents of all five children had normal platelet counts. In addition, there was no family history suggestive of thrombocytosis or haemorrhagic or thrombotic events. Neither of two siblings had increased platelet numbers.

Three out of the five patients had a slightly increased spleen volume at ultrasonographic examination, while no palpable spleen was felt.

At the time of diagnosis, the platelet counts of our patients ranged between 1112 and 3178×10^9/l (mean $1593 \pm 913 \times 10^9$/l) and remained persistently high over the follow-up period. No alteration in white blood cell (WBC) or haemoglobin level was observed. Cytochemical reaction for leucocyte alkaline phosphatase was normal in all patients.

Bone marrow aspirate revealed increased numbers of megakaryocytes with normal features at both mature and immature stages. All other haemopoietic series were present with normal distribution and morphology. The iron content (Pearl's reaction) in bone marrow was normal. Trephine biopsy confirmed these findings and also showed an absence of marrow fibrosis (reticuline staining). In all five patients, classic cytogenetic analysis was normal, and molecular study failed to detect bcr-abl rearrangement.

Table I. Main characteristics of children with essential thrombocythaemia.

Patient	Age at diagnosis	Sex	Follow-up (months)	Symptoms	Treatment	Platelets ($\times 10^9$/l)	WBC ($\times 10^9$/l)	Hb (g/dl)	EPO (U/l)	TPO (pg/ml)	5-HT nM/10^9 platelets	Aggregation		
												ADP	Adr	Coll
A	9 months	F	36	Inferior cava vein thrombosis	Warfarin (INR 2–3)	1112	11·8	12·1	16	59·8	0·93	N	N	N
B	7 years 7 months	F	195	None	None	1200	10·01	13·7	10	30·14	0·46	0	0	N
C	10 years 9 months	F	54	None	None	930	10·2	11·9	11·7	26·87	1·1	N	N	N
D	7 years 9 months	F	121	Headache	ASA + IFN	3178	9·8	1·2	8·3	–	0·94	N	N	N
E	10 years 3 months	M	111	Headache, epistaxis	ASA	1548	11·93	13·5	10	18·27	0·43	N	0	N
Normal values						150–450	4·2–12·4	131–163	11·2 ± 3	35 ± 22	1·5–3	52 ± 14	60 ± 11	70 ± 18

WBC, white blood cells; Hb, haemoglobin; EPO, erythropoietin; TPO, thrombopoietin; 5-HT, intraplatelet serotonin; ADP, adenosine diphosphate; Adr, adrenaline; Coll, collagen; N, normal; 0, absent.

None of the patients had spontaneous BFU-E formation or altered levels of EPO (11·2 ± 1·94 U/l). Platelet aggregations were completely normal in three out of the five patients. Two children showed absent aggregation under adrenaline stimulation. In one case, this was associated with absent ADP-induced aggregation. All patients showed decreased levels of 5-HT (0·77 ± 0·31 nM/10^9platelets) Serum TPO levels were within the normal range (36·07 ± 21·39 pg/ml) in all four cases evaluated.

DISCUSSION

Essential thrombocythaemia is a myeloproliferative syndrome characterized by persistent thrombocytosis for which no aetiology can be determined. Its clonal origin has been established in most cases (Fialkow *et al*, 1981). Thrombocytosis is also observed in other chronic myeloproliferative disorders or may be reactive to many conditions (Murphy, 1983). The distinction between ET and reactive thrombocytosis (RT) is clinically relevant because thrombohaemorrhagic complications are more common in ET. Moreover, in rare cases, ET can transform into acute leukaemia or myelofibrosis (Randi *et al*, 1999b). Therefore, prognosis and management are quite different.

Thrombocytosis in childhood is not rare, as it occurs in 6–15% of paediatric patients (Sutor, 1995). However, ET is extremely rare in childhood, and the cases reported here, as far as we know, represent the largest group followed by the same department (Michiels & Van Genderen, 1997). Moreover, there has been only one case of ET described in an infant (Kapoor *et al*, 1996). Our 9-month-old patient is therefore the second case reported in this age group.

All our patients met the diagnostic criteria of the PVSG for adult ET and, in particular, no cause of reactive thrombocytosis was ascertained. On this basis, a diagnosis of ET could be established in all of them.

However, laboratory and clinical features found in our patients were not always consistent with other data from the literature.

First, chromosomal abnormalities, especially 20q deletions and 13q deletions, have been described in patients with ET, but no consistent chromosomal changes have been observed in adult patients (Green, 1999) as well as in our cases. Moreover, BFU-E-derived colony formation from peripheral blood, in a serum containing medium without the addition of burst-promoting activity or erythropoietin (unstimulated BFU-E), has been observed in most ET patients. Unstimulated BFU-E yield was significantly greater in ET than in RT and controls (Eridani *et al*, 1987), while none of our patients presented unstimulated BFU-E. Mild increases in serum EPO have been observed occasionally in ET patients (Bourantas *et al*, 1995), but not in the five children reported here. Finally, serum TPO was within normal limit, while in ET patients, TPO is usually increased compared with levels in normal subjects (Griesshammer *et al*, 1998) and with patients with reactive thrombocytosis (Hou *et al*, 1998).

On the other hand, the finding of a mild splenomegaly, as observed in three cases only by ultrasonographic scanning, is in agreement with thrombocytosis of clonal disorder. However, splenic enlargement seems to be significant when documented by radionuclide scan (Dudley *et al*, 1989). Of course, this was not performed to avoid exposure to radiation. Moreover, platelet functional alterations have been described in most adult patients with increased platelet counts. A variety of altered platelet aggregation patterns have been observed in thrombocytosis resulting from myeloproliferative disorders (Fabris *et al*, 1984). Absent aggregation under adrenaline stimulation, as we have documented in two out of five children, is probably the most common alteration observed. Low levels of intraplatelet 5-HT seem to be correlated with a thrombocytosis resulting from myeloproliferative disorders (Fabris *et al*, 1984). In agreement with data from the literature, our patients also presented with low levels of 5-HT.

In adults with ET, thrombosis is the most common and hazardous complication, occurring in about 40% of patients (Randi *et al*, 1991). In contrast, most children with ET seem to have few major thrombotic complications (only one patient of the 11 cases reviewed by Michiels & Van Genderen, 1997), but one out of our five patients was found because of a major vein thrombosis. It is also remarkable that, in four patients, extreme thrombocytoses (over 1500×10^9/l) were observed, but no haemorrhages occurred.

Administration of IFN, which has been suggested as a treatment for ET in adults, was tried in one of our patients, without achieving either a remarkable or a lasting reduction in platelet count. The possible complications of IFN therapy (Steegman *et al*, 1998) must be taken into account when deciding on treatment for these patients.

No transformation into acute leukaemia or myelofibrosis has been observed in our children to date.

In spite of the diagnosis of ET strictly according to PVSG criteria, the features discussed do not suggest the presence of a myeloproliferative disorder or, more specifically, of ET but rather of an ‘idiopathic thrombocytosis’ without myeloproliferation. In the past, other possible variants of ET, particularly common in young adults, have been described, such as young people with cerebral sinuses thrombosis (Randi *et al*, 1994) and patients with atypical myeloproliferative disorder with high thrombotic risk but slow disease progression (Barosi *et al*, 1991). Non-clonal ET has also been reported recently in adult patients with a low risk of thrombotic events (Harrison *et al*, 1999). The heterogeneous nature of ET is also suggested by our paediatric patients.

In keeping with the observation of familial cases of polycythaemia (De La Chapelle *et al*, 1993), abnormalities of the TPO receptor or of its regulatory pathway (Yoshida *et al*, 1998) could be considered as causes for increased platelet numbers. In fact, TPO levels were normal in all the cases tested. Therefore, studies are needed to identify such possibilities, even if they do not seem to apply to adult ET (Taksin *et al*, 1999).

Only a careful, continuous observation of these rare cases will elucidate the issues concerning their clinical aspects and therapeutic options.

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