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# Polycythemia Vera

## I. Histopathology, Ultrastructure and Cytogenetics of the Bone Marrow in Comparison with Secondary Polycythemia*

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**Summary.** Clinical and morphological studies including light microscopy, electron microscopy and karyotyping of the bone marrow, were performed on a total of 164 patients with polycythemic conditions. A final diagnosis was obtained from clinical findings and histopathology of plastic embedded core biopsies of the bone marrow including sequential examinations. 51 patients revealed a secondary polycythemia whereas 113 displayed polycythemia vera (P. vera). In this last group 83 cases have persisting P. vera. 30 showed a transgression towards chronic myeloid leukemia with or without accompanying myelofibrosis – osteomyelosclerosis (so called chronic megakaryocytic-granulocytic myelosis – CMGM).

The histopathology of the bone marrow in P. vera revealed consistent alterations which are useful in distinguishing this disorder from secondary polycythemia (SP) and CMGM: depletion of iron storage, increased neutrophilic granulopoiesis but no gross atypia in maturation, polymorphism of megakaryocytes with conspicuous giant forms and dilatation and increased branchings of venous sinusoids. Electron microscopic findings were in agreement and showed further abnormalities of cytological maturation in the erythrocytic and granulocytic lineage. Cytogenetic studies in 27 non-treated patients with P. vera revealed the Philadelphia chromosomes in 2 cases, whereas in SP only minor chromosomal anomalies have been encountered in a few patients.

It is concluded that histopathology of trephine biopsies of the bone marrow is an invaluable aid to establish a correct diagnosis, differentiating P. vera from the other potentially polycythemic disorders and helping to detect a possible progression towards leukemia at an early stage. Cytogenetic investigations may show early structural and numerical abnormalities of the karyotype and possibly precede a presumptive transgression towards

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myeloid leukemia (CMGM). A simultaneously performed histological and chromosomal examination of bone marrow samples is therefore desirable in each case of a polycythemic condition.

**Key words:** Polycythemia vera – Secondary polycythemia – Histopathology – Ultrastructure – Cytogenetics – Bone marrow biopsy.

## Introduction

The diagnosis of a polycythemic disorder may confront the clinician with some problems, particularly in the differentiation of polycythemia vera (P. vera) from secondary polycythemia (reviews by Wasserman 1976; Ellis and Peterson 1979). For this reason hematological data should be supported by histopathology of the bone marrow from core biopsies since smears of marrow aspirates are widely thought to be inadequate for the distinction of P. vera from other polycythemic conditions (Roberts et al. 1969; Duhamel et al. 1970; Lennert et al. 1975). In addition to the histopathology of the bone marrow a simultaneously performed cytogenetic investigation seems to be promising with regard to the various chromosomal anomalies which have been reported in P. vera by Wurster-Hill and McIntyre (1978). These aberrations of the karyograms should not be encountered in patients with a reactive or secondary polycythemia. A combined morphological investigation by light- and electron microscopy was therefore performed together with a cytogenetic study in patients with polycythemia. The aim of this study was to find out whether there exist characteristic features of the fine structure of the bone marrow in P. vera and whether these are accompanied by chromosomal anomalies.

## Patients and Methods

*Patients.* A total of 164 patients with the suspected clinical diagnosis of P. vera have been referred to the hematological service of the Medical School Hannover and various university affiliated hospitals nearby (see acknowledgements) over the last 8 years. A final diagnosis was established from the clinical findings (following the parameters of the Polycythemia vera-Study Group; Berlin 1975; Wasserman 1976), the course of disease and bone marrow biopsies.

*Methods.* Biopsies of the bone marrow were performed as trephines from the iliac crest following the methods of Burkhardt (1966) or Jamshidi and Swaim (1971). Further processing of resin embedding, semithin sections and staining techniques (Giemsa, Gomori's silver impregnation, Goldner's trichrome, methyl-green-pyronin and Prussian blue reaction) were done using routine methods by Burkhardt (1971). Preparation of bone marrow tissue for electron microscopy has been reported in detail elsewhere (Thiele et al. 1977).

Cytogenetic methods included direct preparations of bone marrow samples obtained from the site of the trephine biopsy. Metaphases were evaluated from short time cultures without phytohemagglutinin (PHA) stimulation. Staining was done using conventional methods and Giemsa-banding technique and chromosomal analysis was performed on photographs or on photographic negatives. To report our findings the nomenclature proposed at the Paris conference 1971 and the criteria established by the Polycythemia vera-Study Group were applied (reviews by Wurster-Hill et al. 1976; Wurster-Hill and McIntyre 1978).

**Table 1.** Survey of patients with polycythemia vera and secondary polycythemia. All final diagnoses were established by clinical findings, course of the disease and biopsy of the bone marrow

| Polycythemic disorder | | |
|---|---|---|
| 134 | | |
| 83 | 51 | |
| Polycythemia vera | Secondary polycythemia | |
| F:M/1:1.3 | F:M/1:32 | Sex ratio |
| 48 | 55 | Mean age (years) |
| 27 $^{32}P$, cytostatics | | |
| | none | Treatment |
| 56 phlebotomy only | | |

## Results

By final diagnosis the total of 164 polycythemic patients had to be divided into 3 groups: Firstly P. vera (83 patients), secondly secondary polycythemia (51 cases) which are listed in Table 1, and finally cases with transgression into myeloid leukemia (30 patients, see also Vykoupil et al. 1980).

The main clinical and hematological findings of a selected fraction of 10 patients with secondary polycythemia and 15 cases with P. vera which had been diagnosed at the hematological service of the Medical School Hannover are given in Table 2. This relatively small group of patients has been chosen to demonstrate principle clinical findings in a homogeneous thoroughly investigated fraction of patients. The remaining cases from outside hospitals have closely comparable hematological data.

Morphological findings by light and electron microscopy from the bone marrow in the polycythemic disorders are shown in the illustrations (Figs. 2a, b – 7a, b) in comparison with a normal specimen (Fig. 1a, b).

In the 51 patients with secondary or reactive polycythemia the bone marrow biopsy showed hyperplasia of erythropoiesis with an apparently normal maturation (Fig. 2a, b). In addition there was a normal amount of siderin deposit in the (histiocytic) reticulum cells of the interstitium. Granulo- and megakaryopoiesis were not remarkably altered (Fig. 2b) and the hyperemic sinuses revealed no increase in number and branchings.

In contrast to this, 83 patients with P. vera displayed a conspicuous hyperplasia of erythropoiesis with an atypical dislocalization towards the osseous trabecula, the original generation zones of granulopoiesis (Fig. 3a). There was, in addition a total lack of iron storage in the reticulum cells and a remarkable dilatation and increased branching and tortuosity of the sinuses (Fig. 3a–c). Ultrastructure confirmed these findings by showing extensive islets of immature erythropoiesis with many early and late normoblasts besides erythroblasts (Figs. 4a, b, 6).

**Table 2.** Principle clinical and hematological data in a total of 25 patients with polycythemic conditions (polycythemia vera and secondary polycythemia, diagnosed and treated at the Medical

| Final diagnosis | No. | Sex (ratio) | Age (years) | Hemoglobin g% | Hematocrit % |
|---|---|---|---|---|---|
| Secondary polycythemia | 10 | F:M/0:10 | $\bar{x}$ 49 | 17.7 | 49.8 |
| | | | Sx 12.0 | 1.4 | 3.0 |
| Polycythemia vera | 15 | F:M/6:9 | $\bar{x}$ 58 | 20.9 | 61.3 |
| | | | Sx 11.2 | 2.3 | 6.1 |

Erythrocyte differentiation was apparently disturbed since erythroblasts were encountered with abnormal lobulations (Fig. 5a) or at least atypical indentations (Fig. 5b) of their nuclei. In later normoblasts the most striking alteration of nuclear fine structure consisted of membrane bound splits of the peripheral chromatin (Figs. 4b, 5c). Surveys of electron microscopical specimens exhibited frequent profiles of extended sinuses in between the large areas of erythro- and granulopoiesis (Fig. 6).

Light microscopy of neutrophilic granulopoiesis demonstrated hyperplasia with a shift to the left but no major disorganization of maturation (Fig. 3a). Megakaryopoiesis was grossly abnormal, because there were conspicuous giant megakaryocytes (Fig. 3b) and some microforms apart from an increase in number and polymorphism with many immature cells (Fig. 3c).

These macromegakaryocytes displayed no obvious disturbances of maturation, but a hyperlobated nucleus with numerous segments connected by chromatin bridges and an extensive area of cytoplasm were often present. In overviews electron microscopy showed those large megakaryocytes to contain an abundance of demarcation membranes, specific granules of the bull's eye type and clusters of glycogen (Fig. 7a). There were also very small forms with non-extensive cytoplasm and paucity of organelles (Fig. 7b).

Cytogenetic findings are listed in Tables 3, 4, where the major chromosomal abnormalities and the ratio of metaphases with such anomalies are given.

In secondary polycythemia in 6 out of 10 patients no chromosomal aberrations were found (Table 3). In 4 cases however, extra chromosomes were noted in sporadic metaphases.

P. vera displayed major cytogenetic anomalies with extra chromosomes, aneuploidy and translocation in 17 of 27 cases (63%) (Table 4). Noteworthy is the finding of the Philadelphia chromosome (Ph′-chromosome) in 2 cases particularly in case 1 (W.R., Table 4): This patient was referred to us in June 1975 about 5 years ago with an evident P. vera, when the Ph′-chromosome was present in 7 out of 92 metaphases and additional chromosomal anomalies of the C and D group chromosomes were also observed. At that time, although

School Hannover, Department of Immunology and Blood Transfusion ($\bar{x}$ mean and standard error Sx of the mean for each variable). ALP = alkaline leukocyte phosphatase score

| Peripheral blood count (cu mm) | | | ALP | Total red cell volumen cc/kg | Plethora (ratio) | Spleno-megaly (ratio) |
|---|---|---|---|---|---|---|
| Erythro-cytes $\times 10^6$ | Leuko-cytes $\times 10^3$ | Thrombo-cytes $\times 10^3$ | | | | |
| 5.4 | 7.4 | 188.5 | 65 | 65 | – | – |
| 0.6 | 2.5 | 51.6 | 41 | 12.4 | | |
| 7.5 | 10.8 | 392 | 197 | 77.5 | | |
| 0.7 | 4.3 | 169.7 | 120 | 105 | +9/++6 | −8/+7 |

cytogenetic studies demonstrated the Ph′-chromosome, there was no evidence clinically or histologically for myeloid leukemia. In the meantime (1980) however, a remarkable rise of the leukocyte and platelet count was seen in the peripheral blood and cytostatic therapy was initiated as early treatment of an anticipated transformation into myeloid leukemia. In the other patients a normal karyotype was noticed in 10 of 27 cases (37%).

## Discussion

About one third of the 164 patients who entered our study of polycythemia actually displayed a secondary or reactive polycythemia (SP). This relatively high proportion of SP in our series may be due partially to the prior evaluation of marrow smears only which are thought to be insufficient for the differential diagnosis of the various forms of polycythemic conditions (Lennert et al. 1975; Roberts et al. 1969). Hence the histology of bone marrow biopsies is an essential requirement to establish a correct diagnosis. The fine structure of hematopoiesis and the myeloid stroma, which are most favorably preserved in plastic embedded specimens allow an easy and clear cut decision. Although the importance of the bone marrow biopsy as the essential diagnostic tool has been debated (Ellis et al. 1975), most regard this method as an invaluable aid (Krasznai et al. 1969; Burkhardt et al. 1969; Duhamel et al. 1970; Kurnick et al. 1972). Examination of biopsies of the bone marrow is not only necessary to establish the diagnosis, but also to follow the course of disease by sequential corings (Duhamel et al. 1970; Burkhardt et al. 1969, 1979). Neither hematological data including the score of the alkaline phosphatase, nor cytogenetic analysis clearly separate every case of P. vera from SP or early stage leukemia (Wasserman 1976; Berlin 1975).

Histopathology of the bone marrow in P. vera shows characteristic features. Differential diagnosis with regard to other polycythemic disorders can be based on reliable criteria which are given in Table 5. Although the histology of

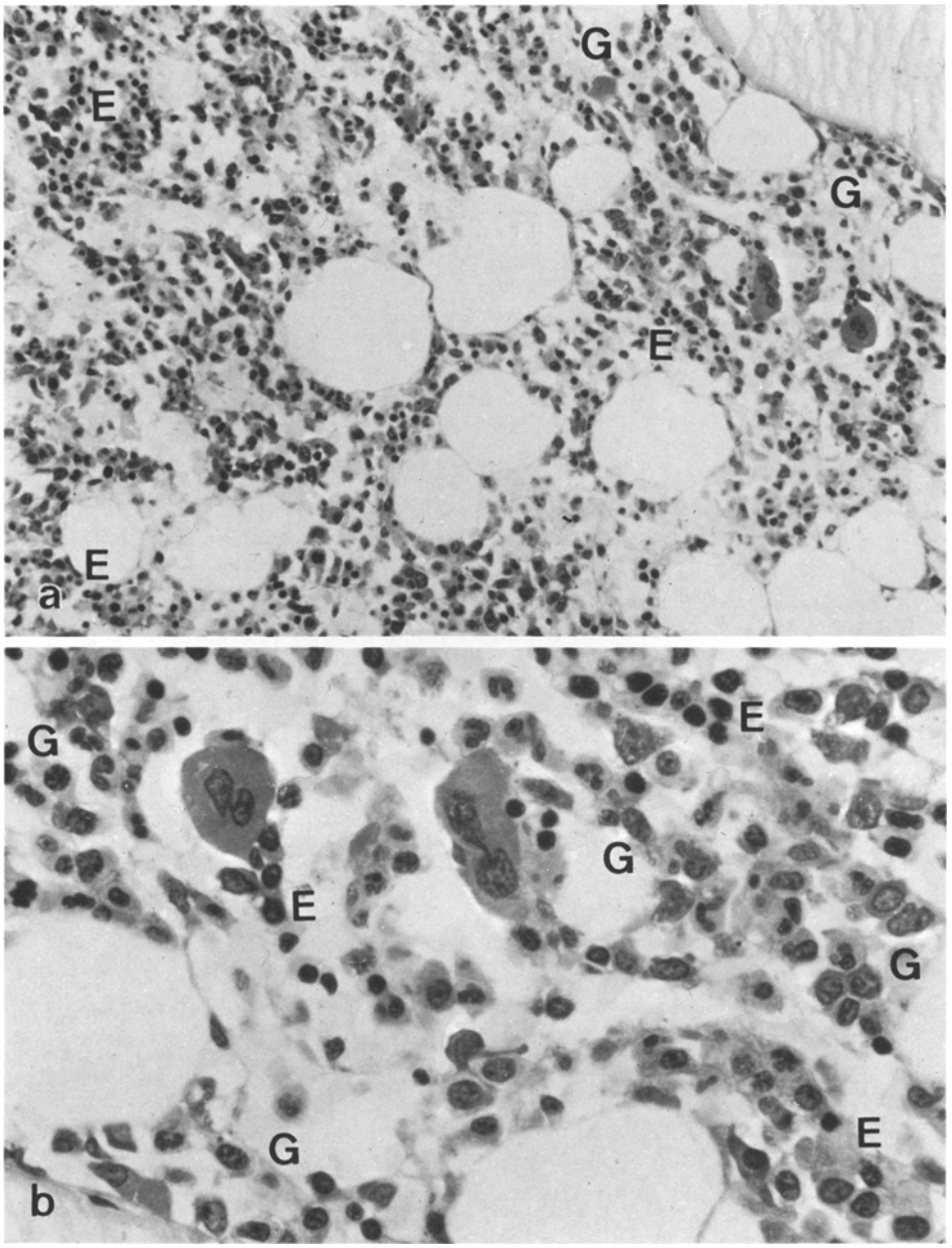


**Fig. 1 a and b.** Normal bone marrow as control. **a** Survey with dispersed islets of erythropoiesis (*E*) and mostly peritrabecular granulopoiesis (*G*). **b** Two megakaryocytes with regularly lobated nuclei and large areas of cytoplasm surrounded by erythrocytic (*E*) and granulocytic (*G*) precursor cells. **a** × 280; **b** × 620

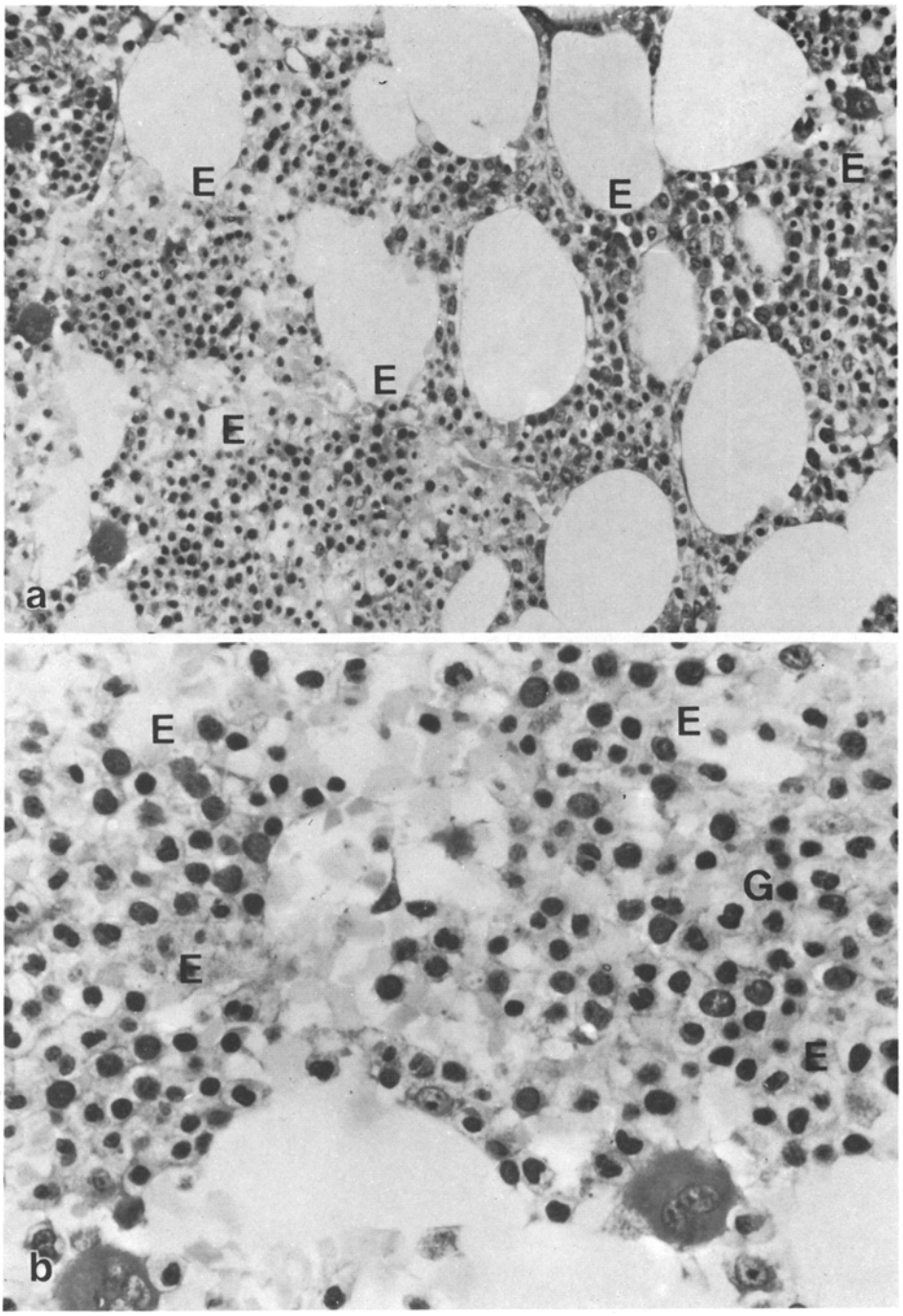


**Fig. 2a and b.** Secondary polycythemia. **a** Survey with extensive islets of erythropoiesis (*E*) containing late and early normoblasts with round dark nuclei; neutrophilic granulopoiesis seems to be almost effaced. **b** Two megakaryocytes with normal appearance are surrounded by large clusters of erythroblasts and normoblasts,cytes (*E*) of different stages of maturation, there are only a few granulocytes (*G*). **a** × 280; **b** × 620

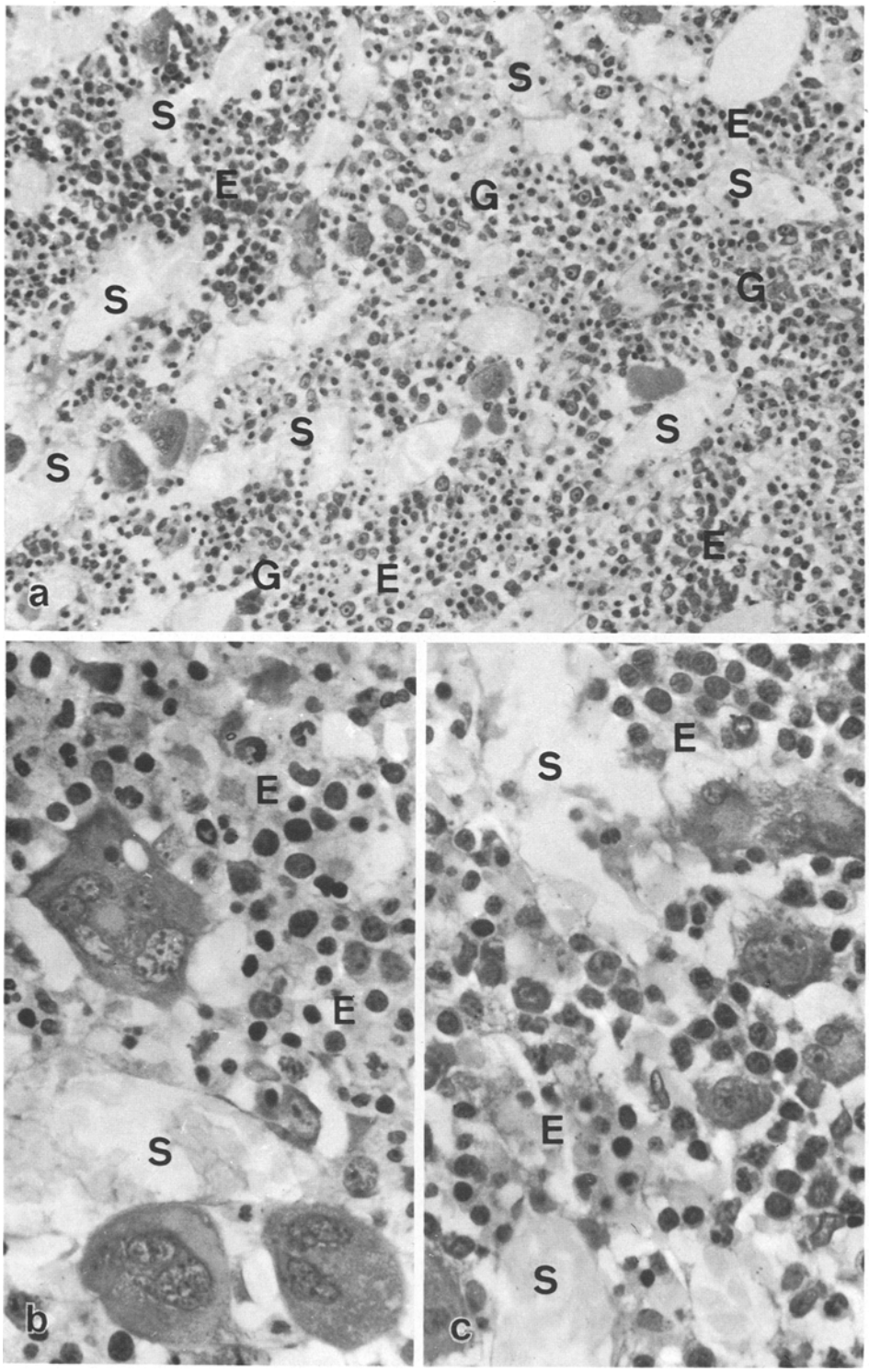


**Fig. 3a–c.** Polycythemia vera (P. vera). **a** Survey with conspicuous hyperplasia of erythropoiesis (*E*) and dilatation and branchings of sinuses (*S*). In between large areas of neutrophilic granulopoiesis with a shift to the left (*G*). **b** Giant megakaryocytes in P. vera with a hyperlobated nucleus and stag horn-like formation of nuclear segments which are connected by chromatin bridges, located along a sinus (*S*) and surrounded by immature erythropoiesis (*E*). **c** Small megakaryocytes and -blasts with only a rimlike area of cytoplasm with nearby sinuses (*S*) and extensive erythropoietic islets (*E*). **a** × 280; **b** and **c** × 620

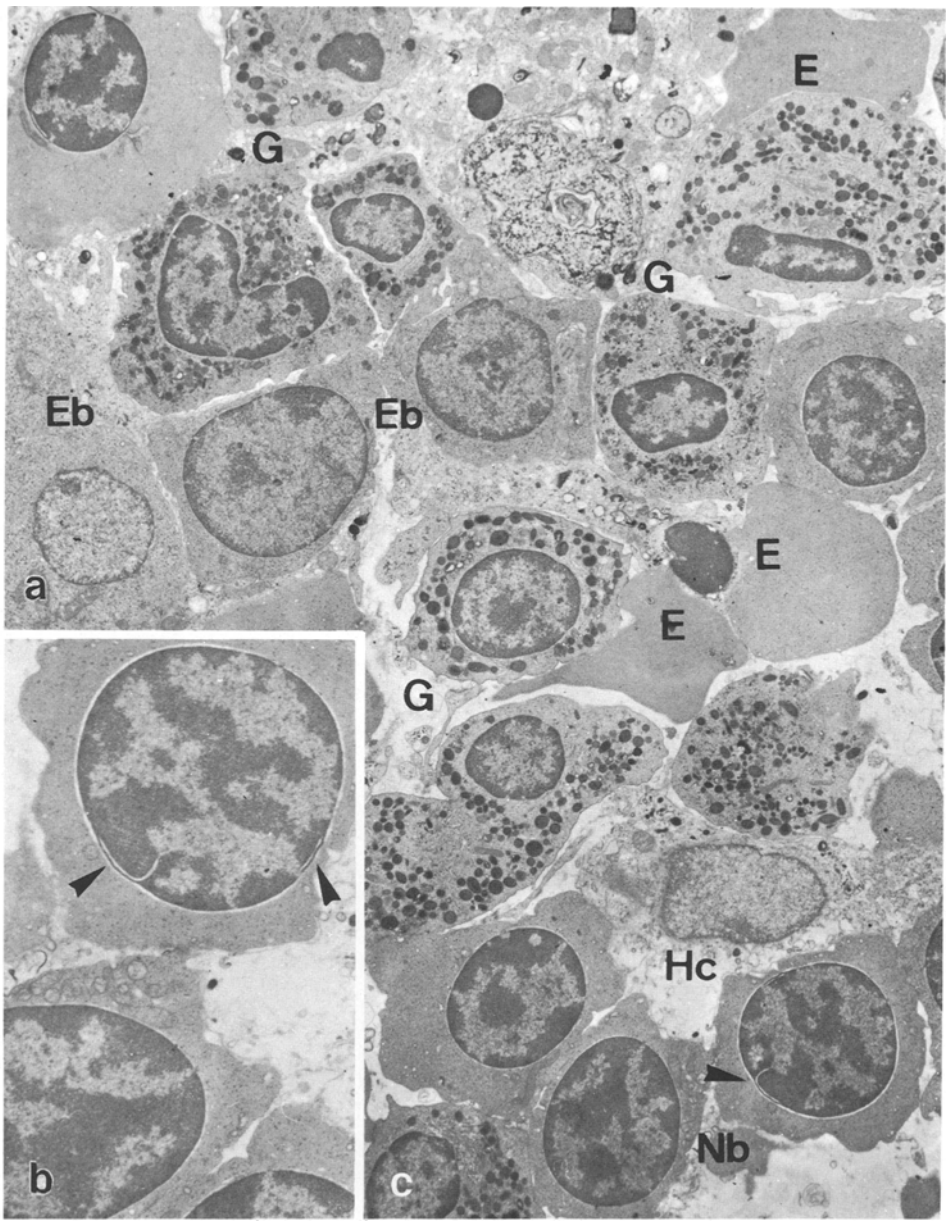


**Fig. 4a and b.** Ultrastructure of P. vera. **a** Survey with erythropoiesis showing late normoblasts (*Nb*), one with a nuclear split (*arrow head*) surrounding a histiocytic reticulum cell (*Hc*), further erythroblasts (*Eb*) and erythrocytes (*E*) besides neutrophilic granulopoiesis (promyelocytes and metamyelocytes, *G*). **b** Late normoblast with two nuclear splits of the peripheral chromatin (*arrow heads*). **a** × 5,000; **b** × 9,000

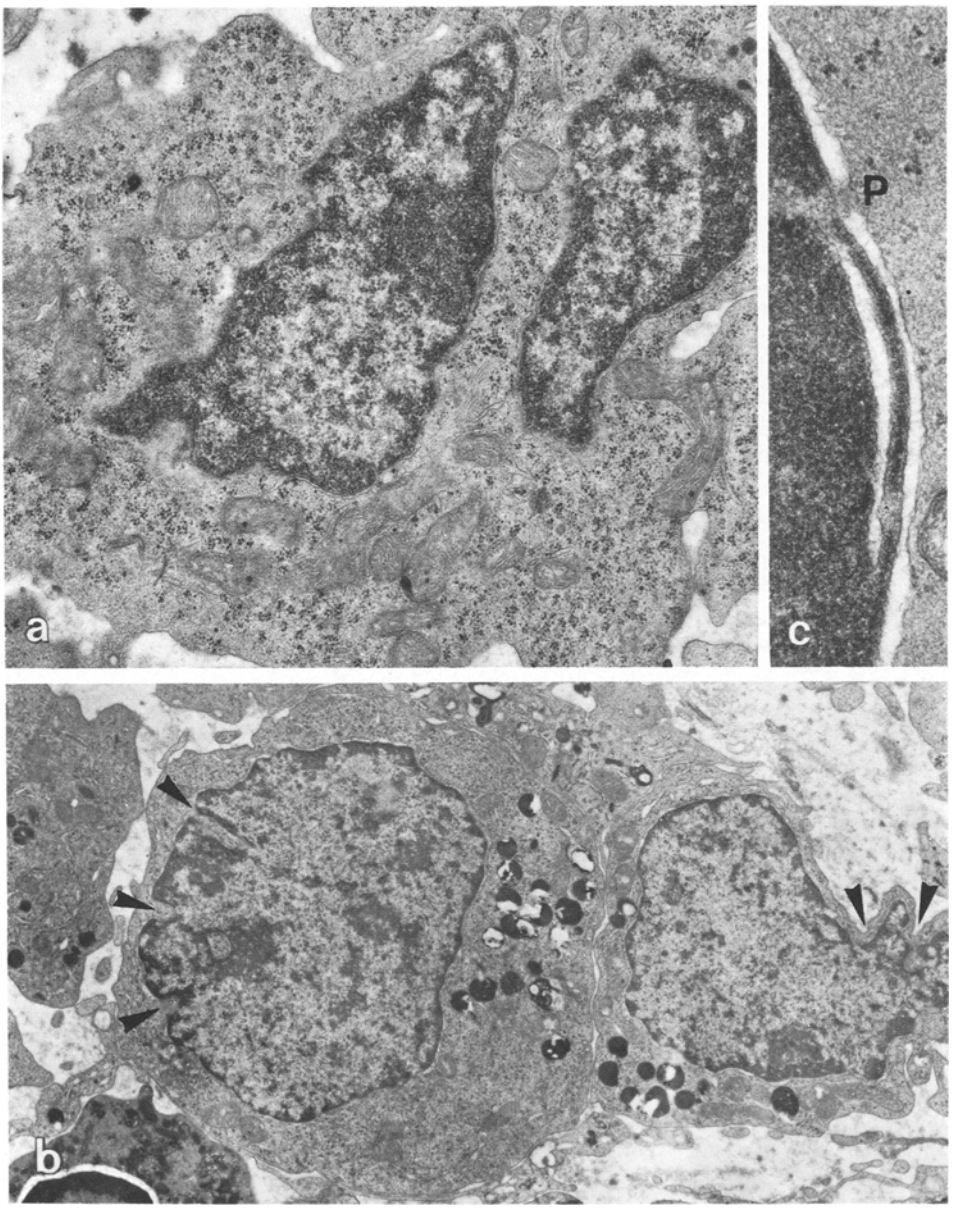


**Fig. 5a–c.** Ultrastructural atypias of erythropoiesis in P. vera. **a** Abnormal lobulation of erythroblast containing many mitochondria and free polysomes. **b** Two late erythroblasts with atypical nuclear indentations (*arrow heads*) and dense siderosome-like bodies in their cytoplasm. **c** Membrane bound cleft of the nuclear chromatin associated with a nuclear pore (*P*). **a** × 18,000; **b** × 10,000; **c** × 45,000

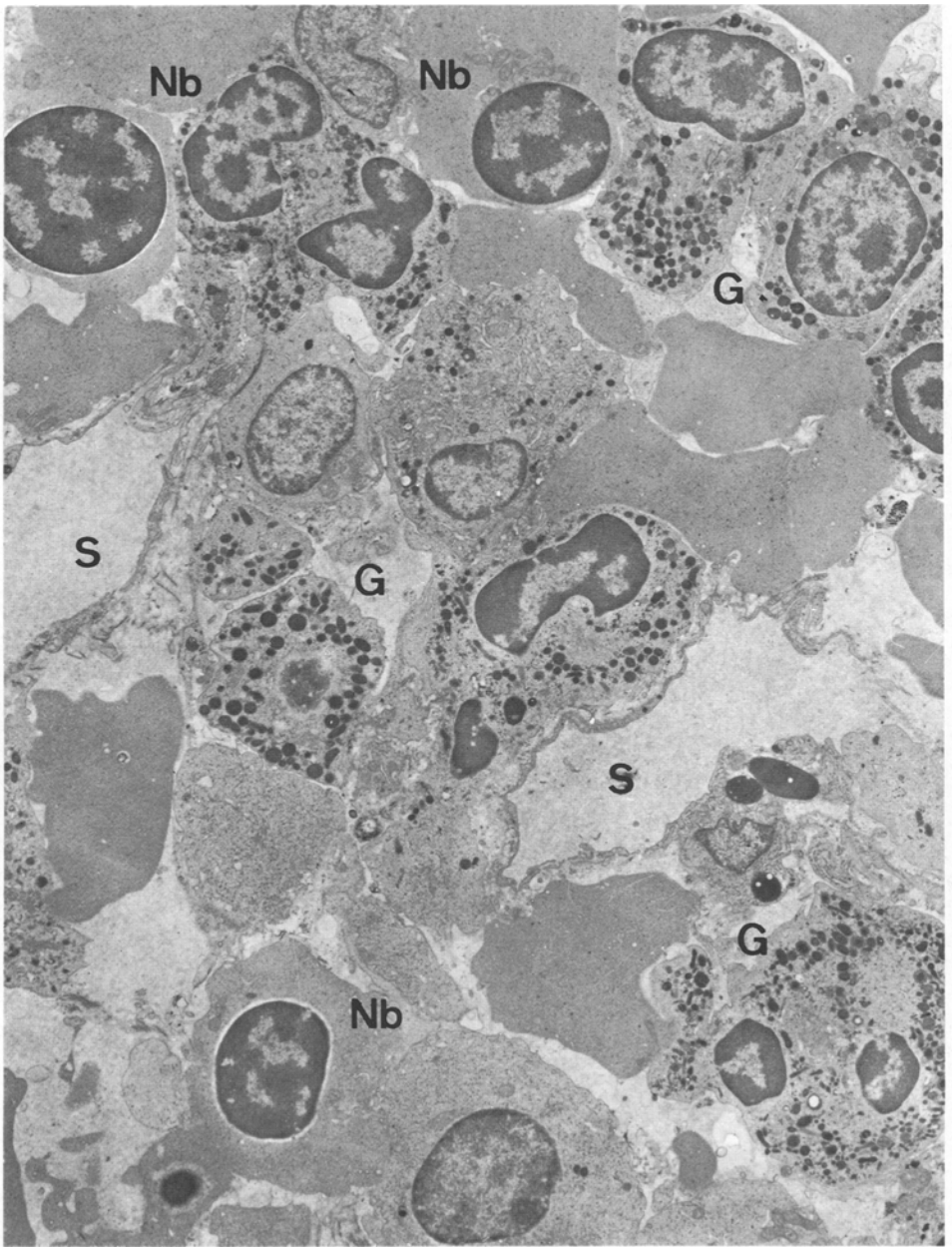


**Fig. 6.** Electron microscopy of P. vera with distended sinus (*S*) surrounded by early and late normoblasts (*Nb*) and granulopoiesis (promyelocytes-metamyelocytes, *G*). × 5,000

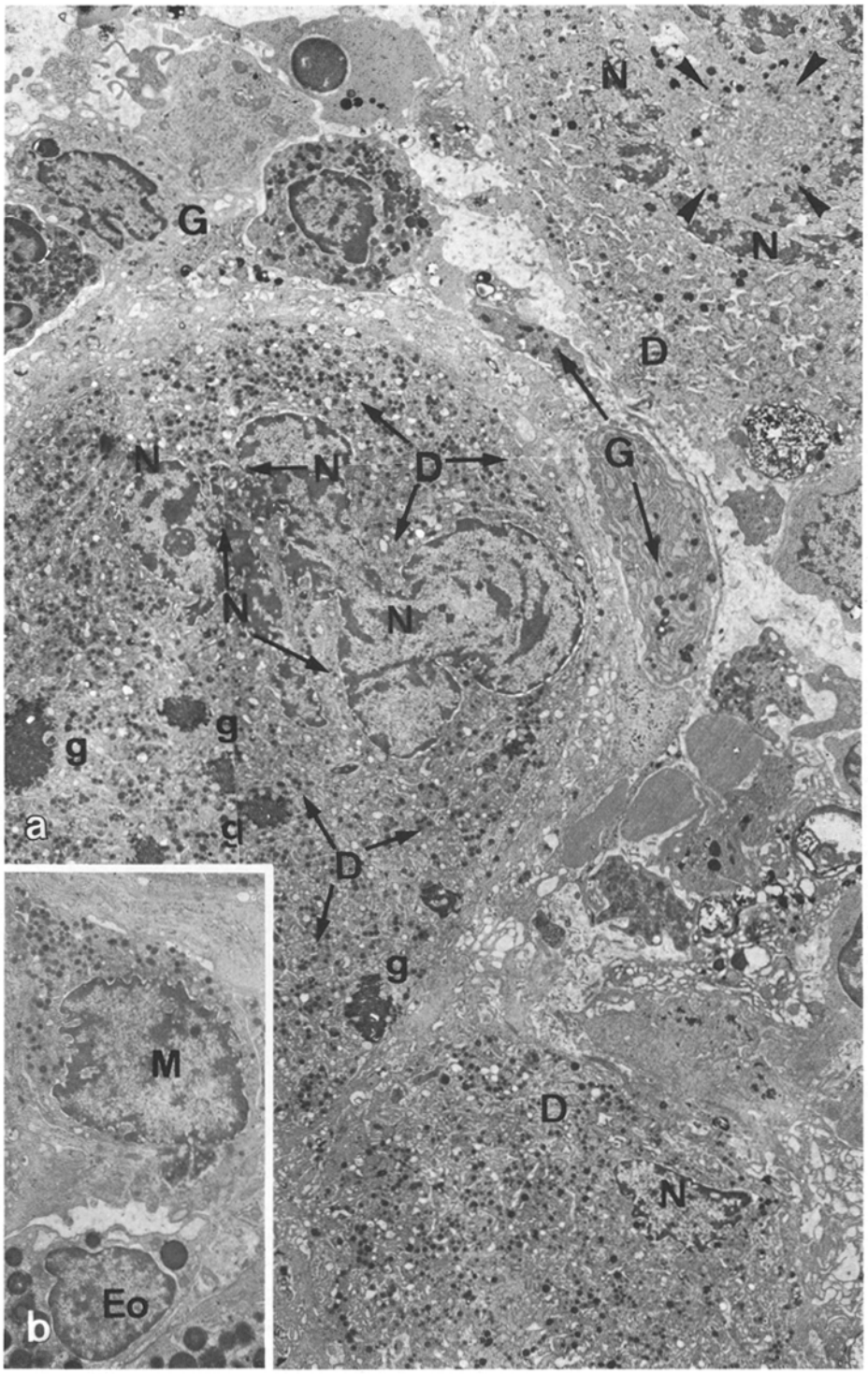


**Fig. 7a and b.** Ultrastructure of megakaryopoiesis in P. vera. **a** Survey with parts of three large megakaryocytes with lobated nuclei (*N*), clusters of glycogen (*g*), abundant specific granules and extended demarcation membrane systems (*D*), sometimes in whorl-like formations (*arrow heads*). **b** Small megakaryocytes (microform: *M*) in comparison with an eosinophilic promyelocyte (*Eo*). There is only a small rim of cytoplasm surrounding an apparently mature nucleus in the micro-megakaryocytes. **a** × 3,500; **b** × 4,000

**Table 3.** Cytogenetic findings in 10 non-treated patients with secondary polycythemia; the patients 7–10 show extra chromosomes in sporadic metaphases

| | Initials | Chromosomal anomalies | Ratio abnormal/ metaphases |
|---|---|---|---|
| 1. | A. E. | – | 0/17 |
| 2. | B. W. | – | 0/17 |
| 3. | H. H. | – | 0/17 |
| 4. | K. W. | – | 0/17 |
| 5. | R. H. | – | 0/32 |
| 6. | T. B. | – | 0/20 |
| 7. | B. W. | +10 | 1/17 |
| 8. | C. S. | +16, +17, +22 | 2/17 |
| 9. | F. A. | +11 | 1/35 |
| 10. | L. G. | +6, +16 | 2/17 |

**Table 4.** Cytogenetic findings in 27 non-treated patients with P. vera at the time of the first biopsy. There are two cases with a Ph′-chromosome and the majority of the other patients shows abnormalities of the C–D group chromosomes, particularly No. 8 (underlined). Case 1 (W.R.) had his cytogenetic investigation done in 1975, since then there is an increasing clinical evidence for an insiduous transgression towards myeloid leukemia.

| | Initials | Chromosomal anomalies | Philadelphia chromosome | Ratio abnormal/ metaphases | | Initials | Chromosomal anomalies | Ratio abnormal/ metaphases |
|---|---|---|---|---|---|---|---|---|
| 1. | W. R. | +8, +10, +12, +16 | + | 18/92 | 14. | W. H. | +8, +11, +22 | 3/16 |
| 2. | Sch. K. | | + | 1/6 | 15. | W. E. | +6 | 2/17 |
| 3. | B. A. | +22 | | 2/35 | 16. | G. W. | +19, +22 | 1/12 |
| 4. | F. M. | +6, 11, +14, +22 | | 3/17 | 17. | H. H. | +12 | 1/13 |
| 5. | F. B. | +5, +8 | | 6/70 | 18. | H. A. | | 0/27 |
| 6. | H. V. | +2, −10 | | 5/34 | 19. | W. F. | | 0/7 |
| 7. | H. W. | +13q, 2+ | | 2/29 | 20. | S. R. | | 0/14 |
| 8. | J. A. | +15, +21 | | 7/69 | 21. | R. A. | | 0/17 |
| 9. | N. H. | ps.-Tripl. | | 2/17 | 22. | B. A. | | 0/13 |
| 10. | R. W. | +2, −10 | | 2/35 | 23. | M. M. | | 0/17 |
| 11. | S. H. | +2, −8, −10, 15 | | 4/17 | 24. | H. A. | | 0/15 |
| 12. | S. S. | (2q+; 6q−) | | 6/35 | 25. | R. F. | | 0/17 |
| 13. | T.W. | +13, 15q−, 14q (4q+; 20q−) | | 20/100 | 26. | H. H. | | 0/17 |
| | | | | | 27. | A. H. | | 0/17 |

**Table 5.** Reliable criteria of histopathology in P. vera, which can not be found in other polycythemic conditions of the bone marrow, i.e., secondary polycythemia and P. vera with transformation into myeloid leukemia. They therefore permit a clear cut distinction between these entities

| Compartment of the bone marrow | Histopathology |
|---|---|
| Erythropoiesis | Overt hyperplasia with atypical localization towards the osseous trabecula |
| Iron storage | Complete sideropenia |
| Neutrophilic granulopoiesis | Shift to the left, but no cellular atypias |
| Megakaryopoiesis | Increase with shift to the left, polymorphy with conspicuous giant forms besides abnormal microforms |
| Myeloid stroma | Dilatation and increased branchings of sinuses, i.e. hyperplasia of venous sinusoids |

P. vera has been described and reviewed by Burkhardt et al. (1969) and Ellis and Peterson (1979) for the "Polycythemia vera-Study Group" our findings warrant the stressing of a few diagnostic peculiarities of the bone marrow alterations.

There are at least three diagnostic clues which may distinguish P. vera from SP: Depletion of iron stores or sideropenia is described as a constant and most reliable finding in P. vera (Burkhardt et al. 1969; Kurnick et al. 1972; Ellis and Peterson 1979).

The occurrence of large sized megakaryocytes (so called giant forms) is a very conspicuous change in cellular differentiation (compare Figs. 1b and 2b with 3b). The appearance of these remarkable and easily recognizable giant megakaryocytes in P. vera has been well established by several methods including microplanimetric studies of smears (Albrecht and Fülle 1974) and sections (Franzén et al. 1961; Lundin et al. 1972; Kutti et al. 1973). These studies of the bone marrow in P. vera have demonstrated significantly higher megakaryocyte size than in controls, reactive polycythemia or chronic myeloid leukemia. These findings were corroborated by cytophotometry where a frequent occurrence of high ploidy cells at 64 N (Lagerlöf 1972; Queisser et al. 1976) was observed in comparison with controls which mostly belonged to the 16 N class (review by Penington 1979). Ultrastructural studies further confirmed this presence of large megakaryocytes, showing cells with an obvious hyperplasia of organelles and increased nuclear lobulation (Lagerlöf and Franzén 1972; Thiele et al. 1979a).

Hyperplasia of sinuses and small capillaries was first emphasized by Burkhardt et al. (1969), probably because his elaborate method of plastic embedding allows a thorough evaluation of an unaltered myeloid stroma. The processing of marrow samples in paraffin, decalcification and various fixatives may result in a blurring of the fine vascular structures so that this point has not been particularly stressed, not even in papers which deal with the histology of P. vera in the bone marrow (review by Ellis et al. 1975). A skilful quantitative evaluation of the hyperplasia of the venous sinusoids in P. vera was performed by Demmler (1974) who demonstrated that in contrast to the normal bone marrow there is a significant increase: 11.7% of the total volumen in P. vera compared with 5.3% in the control specimen.

Hypercellularity of the bone marrow may be a major feature as reported by Kurnick et al. (1972), Roberts et al. (1969) and Lundin et al. (1972), but a normocellular specimen may not entirely exclude this disease (Ellis et al. 1975; Ellis and Peterson 1979).

Cytogenetics (Table 1) show that 10 patients (out of 51 cases) with clinically suspected P. vera (Table 1) have secondary polycythemia according to the histopathology of the bone marrow and hematological data (Table 2). Although the number of metaphases with abnormal karyotypes is very few, this finding in 4 patients may indicate that this group still contains some early cases of P. vera which may not have been detected by laboratory or histological methods at the time of our investigation. Thus, these chromosomal aberrations are suggestive and herald the possible onset of a P. vera. They need further evaluation in follow-up studies.

It is true that chromosomal abnormalities seen in direct preparations of the bone marrow have been found in cases without clinical evidence of hematological disorders, but as follow-up-studies have demonstrated, many of such patients later developed malignancies. Sometimes there is a long gap, as in one case discussed by Rowley (1976), where two and a half year elapsed between the initial chromosome study and overt manifestation of leukemia. The question of whether a chromosomal abnormality should be used as the only diagnostic clue for leukemia or even so called preleukemia, as was shown for the Ph'-chromosome by Thiele et al. (1979 b) is not resolved. Evidence is gathering, however, which suggests that chromosomal abnormalities point to an impending malignancy at least, i.e. in case 1 (W.R.) with P. vera (Table 4). Of the 27 patients with apparent P. vera all but 10 showed major chromosomal abnormalities before therapy. Since none of these cases received any chemotherapy or radioactive phosphorus ($^{32}P$) prior to the chromosome study, these aberrations could be regarded as inherent to P. vera. This result gives 63% abnormality in comparison to an estimated 26% of non-treated cases (review by Wurster-Hill and McIntyre, 1978). Chromosomal anomalies in P. vera have been found repeatedly by various investigators: Millard et al. (1968); Reeves et al. (1972); Hsu et al. (1974); Rowley (1976) and others (reviews by Kay et al. 1966; Lawler et al. 1970; Wurster-Hill et al. 1976; Westin et al. 1976). Banding studies demonstrated that an extra *C-group-chromosome* (trisomy) may occur which is almost always a No. 8 or 9 (Westin et al. 1976), an anomaly encountered in only three of our patients (cases 1, 5, 14; Table 4). This seems to be a nonspecific change (Wurster-Hill and Maurer 1978).

Special interest was paid to the *F-group*, particularly No. 20 anomalies, since it has been postulated that a deletion of the long arm of No. 20 (20q-) is frequently seen in patients treated with $^{32}P$, irradiation or busulfan (Kay et al. 1966; Weinfeld et al. 1977). The possibility was discussed (Rowley, 1976) that the No. 20 chromosome has a particularly susceptible site for mutagenic actions in P. vera and therefore $^{32}P$ and other therapeutic agents selectively affect this chromosome. Reeves et al. (1972) and Nowell and Finan (1978) found a similar anomaly in treated and non-treated patients. The discussion continues on whether chromosomal abnormalities including specific anomalies of No. 20 or an other aberration of F-group chromosomes are primary or secondary

events in P. vera. Structural abnormalities of the 13–20 group chromosomes (review Wurster-Hill and McIntyre 1978), although described in a number of cases with P. vera, are not specific for this condition, having been observed frequently in various hematological disorders. The Ph′-chromosome has been rarely reported to be present in P. vera, mostly in patients with previous treatment (Kemp et al. 1964; Modan et al. 1970; Verhest and van Schoubroeck 1973). Two patients (cases 1, 2; W.R. and Sch.K., Table 4) showed the Ph′-chromosome in addition to other numerical anomalies prior to therapy. Appearance of the Ph′-chromosome clearly implies a transgression towards malignancy or precedes a leukemic stage of hematological disorder respectively (Rowley 1976). This is confirmed by the further clinical history of case 1 (W.R., Table 4), a patient who after about 5 years of Ph′-positive P. vera now displays clinical findings of an evolving myeloid leukemia. Consequently the value of cytogenetic studies is emphasized in all P. vera patients for the early detection of this trend towards malignancy, as indicated by this specific chromosomal anomaly.

Our results demonstrate that the diagnosis of P. vera should largely depend on the histopathology of the bone marrow particularly with regard to the distinction from reactive polycythemia. Further, chromosomal anomalies may be encountered in patients with SP and P. vera which are thought to herald the onset of P. vera or myeloid leukemia respectively. In conclusion the value of histopathology and simultaneously performed cytogenetic investigations of the bone marrow are emphasized in all polycythemic disorders.

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