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Patho-Anatomical Features of So-Called Ph^{1-} Chronic Myeloid Leukemia

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Summary. Chronic myeloid leukemia without the Philadelphia chromosome (Ph^{1-} CML)* is described and distinguished from chronic myeloid leukemia with the Philadelphia chromosome (Ph^{1+} CML) on the basis of clinical and autopsy findings of four cases. Ph^{1-} CML showed clinical, hematological, and patho-anatomical features which could be regarded as typical.

Patho-anatomically Ph^{1-} CML differed from Ph^{1+} CML in the variable maturation of the leukemic proliferation in the bone marrow and extramedullary infiltrates. Up to the terminal phase Ph^{1-} CML can be of an extremely mature cell type. However, it can also show myeloblastic transformation after an initially mature cell stage. Ph^{1-} CML infiltrates are found in tissues and organs which Ph^{1+} CML usually does not infiltrate or only to a low degree until a blastic crisis.

On the basis of its course and clinical and patho-anatomical features Ph^{1-} CML looks like an atypical chronic myeloid leukemia. However, it is better called an acute myeloid leukemia of the mature cell type.

Key words: Ph^1 negative chronic myeloid leukemia — Ph^1 positive chronic myeloid leukemia — Blood picture, bone marrow, and extramedullary leukemic infiltrates — Pattern of leukemic infiltration of organs and tissues — Myelofibrosis — Patho-anatomical features.

Zusammenfassung. An den klinischen und autoptischen Befunden von vier eigenen Fällen wird das Erscheinungsbild der Ph^{1-} chronischen myeloischen Leukämie (Ph^{1-} CML) dargestellt und gegenüber der Ph^{1+} CML abgegrenzt. Ph^{1-} CML weisen klinische, hämatologische und pathologisch-anatomische Besonderheiten auf, die bei jedem atypischen Verlauf an das Vorliegen des von uns beschriebenen Typs denken lassen sollten.

Pathologisch-anatomisch differiert die Ph^{1-} CML von der typischen CML, der Ph^{1+} CML, durch die von Fall zu Fall wechselnd weite Ausreifung der myeloischen Proliferation im Knochenmark und in den extramedullären Infiltraten wie auch durch die Verteilung der Infiltrate in Organen und Geweben. Die Ph^{1-} CML kann bis in das Terminalstadium extrem reifzellig sein, aber auch nach initial reifzelligem Bild eine myeloblastäre Transformation zeigen. Zudem siedelt sich die Ph^{1-} CML dort an, wo sich die (typische) Ph^{1+} CML in der Regel erst im Blastenschub etabliert oder stärker manifestiert.

Die Ph^{1-} CML imponiert nach Krankheitsverlauf und Erscheinungsbild als eine atypische chronische myeloische Leukämie, sollte aber besser als eine reifzellige akute myeloische Leukämie bezeichnet werden.

The Philadelphia chromosome (Ph^1) is detectable in myelopoietic, erythrocytopoietic, and megakaryocytopoietic cells in more than 85% of the cases of typical chronic myeloid leukemia (Sandberg *et al.*, 1966; Whang-Peng *et al.*, 1968; Ezdinli *et al.*, 1970). Chronic myeloid leukemia (CML) without the Ph^1 chromosome (Ph^{1-} CML) shows particular hematological and clinical features

* Chromosome tests performed by M.-E. Tolksdorf, Department of Pediatrics (Head: Prof. Dr. H.-R. Wiedemann), University of Kiel.

(Krauss *et al.*, 1964; Hardisty *et al.*, 1964; Hellriegel, 1968). It also shows patho-anatomical characteristics which differ from those of Ph^{1+} CML. This will be demonstrated here, since there have been only few patho-anatomical papers dealing with Ph^{1-} CML (see Takanashi, 1972).

Material

Four men, 53 to 83 years old (Table 1). All four patients complained of a deterioration in their general condition starting up to one year before hospitalization. Retrospectively, only one of the patients had a preleukemic phase of four years.

Hematological Findings

Before therapy the leukocyte counts of all 4 patients were less than 50000 cells/μl and terminally above 190000/μl in only 2 cases. The blood smears of two patients showed a mature cell picture (stab forms and polymorphonuclear neutrophils; Cases 1 and 2). Two patients had a mixed mature-immature blood cell picture with 25% myelocytes, including promyelocytes and so-called myeloblasts (about 1% each); terminally the percentage of blasts increased in only one case (Case 3) from 5.5% to 46%. None of the patients had basophilia of more than 1%, none had eosinophilia. In the sternal bone marrow there was massive myeloproliferation. In one case (Case 2) a moderate myelofibrosis could be detected in a needle biopsy from the iliac crest. All patients were anemic on admission—one patient (Case 4) only slightly (4.3×10^6 erythrocytes/μl), one patient (Case 2) severely (1.2×10^6 erythrocytes/μl). All patients had low thrombocyte counts; the highest of all counts was 78000/μl! On admission two of the four patients showed no palpable enlargement of the spleen or liver. Initially, the lymph nodes were not enlarged. Clinically, lymph node enlargement developed in only one patient (Case 3) together with terminal blastic crisis. At autopsy all patients had hepatosplenomegaly and enlarged lymph nodes.

The Ph^1 chromosome could not be found in any of our patients. In 3 of our 4 cases the HbF was not elevated above 5%. The neutrophilic granulocytes of all patients revealed an increased alkaline phosphatase activity.

Three of the 4 patients were cytostatically treated with busulfan; the response to this therapy was poor.

Autopsy Findings

All 4 patients showed signs of hemorrhagic diathesis: cutaneous and mucosal bleeding (Cases 1, 2, 3), hemorrhages in the cerebellum and subarachnoid space (Case 1), chronic subdural hematoma (Case 2), partly massive hemorrhages in the lungs (Cases 2, 3), and in one case (Case 4) massive bleeding in the gastrointestinal tract.

Organs

1. Bone Marrow. Gray-red, in one case (Case 2) dark brown-red bone marrow in all vertebras, in the sternum, iliac crest, and in the whole femoral cavity. Histological findings: Severe myeloproliferation. There were no fat cells (Fig. 1). The number of erythrocytopoietic cells was moderately to strongly reduced in all cases, the number of megakaryocytopoietic cells greatly diminished in Cases

Table 1. Clinical, hematological, and autopsy findings for 4 patients with Ph¹⁻ chronic myeloid leukemia

Case	Age yrs.	Sex	Interval I[a]	Interval II[b]	Leukocyte counts cells/μl[c]	Blood picture[c]	Highest thrombocyte count cells/μl	Enlargement of[c, d]		
								liver	spleen	lymph nodes
1	83	♂	1 (4) yr.	8 weeks	a) 31400 b) 78600 c) 78600	a) predominantly mature (numerous pseudo-Pelger cells) b) mature	78000	a) 10 cm b) 10 cm (2080 g)	6 cm 6 cm (720 g)	Ø Ø (3 × 2 × 1 cm)
2	68	♂	5 mos.	8 weeks	a) 27000 b) 196000 c) 196000	a) predominantly mature b) immature-mature (5.5% blasts)	51000	a) 8 cm b) 8 cm (2600 g)	8 cm 8 cm (400 g)	Ø Ø (3 × 2 × 1.5 cm)
3	53	♂	1 yr.	10 mos.	a) 8088 b) 207000 c) 207000	a) immature b) immature (46% blasts)	62000	a) Ø b) 7 cm (2755 g)	Ø 15 cm (2125 g)	Ø + (3 × 2 × 2 cm)
4	67	♂	a few mos.	10 mos.	a) 44900 b) 64800 c) 64800	a) predominantly mature b) immature-mature (5.5% blasts)	70000	a) Ø b) 10 cm (3210 g)	Ø 4 cm (700 g)	Ø Ø (3 × 2 × 1 cm)

[a] Interval I = amount of time between first symptoms and diagnosis.
[b] Interval II = amount of time between diagnosis and death.
[c] a) initial, b) terminal, c) highest.
[d] Weight or size at autopsy in parentheses.

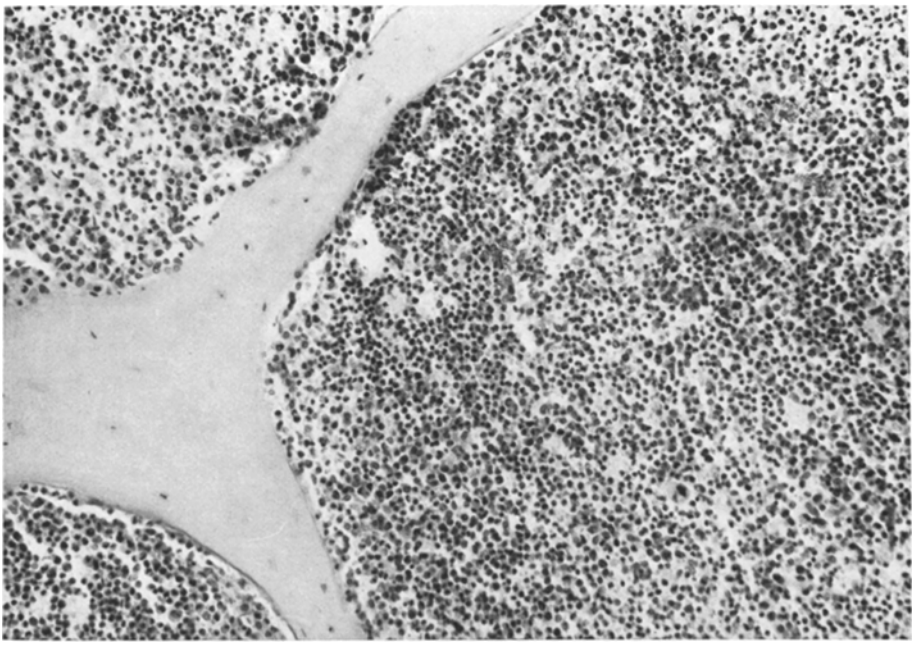

Fig. 1. Vertebra. Strong leukemic infiltration of the bone marrow, rich in mature myelocytic cells. Erythropoietic and megakaryocytopoietic cells greatly reduced in number. No fat cells. 68 year old man (Case 2). H & E, ×140

2 and 4. Cases 1 and 3 showed a normal megakaryocytopoiesis. The granulocytopoiesis of Cases 1, 2 and 4 was mixed mature-immature with prevalence of myelocytes and metamyelocytes and with a high percentage of monocytes in Case 4. Case 3 was rich in naphthol-AS-D-chloroacetate-esterase negative myeloblasts and predominantly immature. The spongiosa was thinned only in Case 2. There was a diffuse increase in the number of reticular fibers in all cases—slight in Case 1, moderate in Cases 3 and 4, and partly great in Case 2 (Fig. 2). Some of the fibers were arranged in a diffuse alveolar network, some in disseminated perivascular and peritrabecular patches or strands (Fig. 7). The sinuses varied from narrow to moderately wide to wide from case to case and occasionally showed perisclerosis (Fig. 8). No case revealed spongiosclerosis or the formation of reticular bone, and there were no precipitates of fibrin and no xanthomatous macrophages.

2. Liver. All patients had at least moderate hepatomegaly (the weights are presented in Table 1: 2080 g, 2600 g, 2755 g, 3210 g). There were macroscopically detectable leukemic infiltrates only in Case 4. Microscopic findings: Strong leukemic infiltration of the periportal spaces—sometimes sharply, sometimes poorly defined (Figs. 3 and 4). On the other hand, the sinusoids were only slightly (Cases 1 and 2) or moderately (Cases 3 and 4) infiltrated. The maturity or immaturity of the leukemic cells in the liver infiltrates roughly corresponded to that of the bone marrow. In the sinusoids there were some macroblasts in Case 2 and some megakaryocytes in Cases 2 and 3. None of the cases showed cirrhosis.

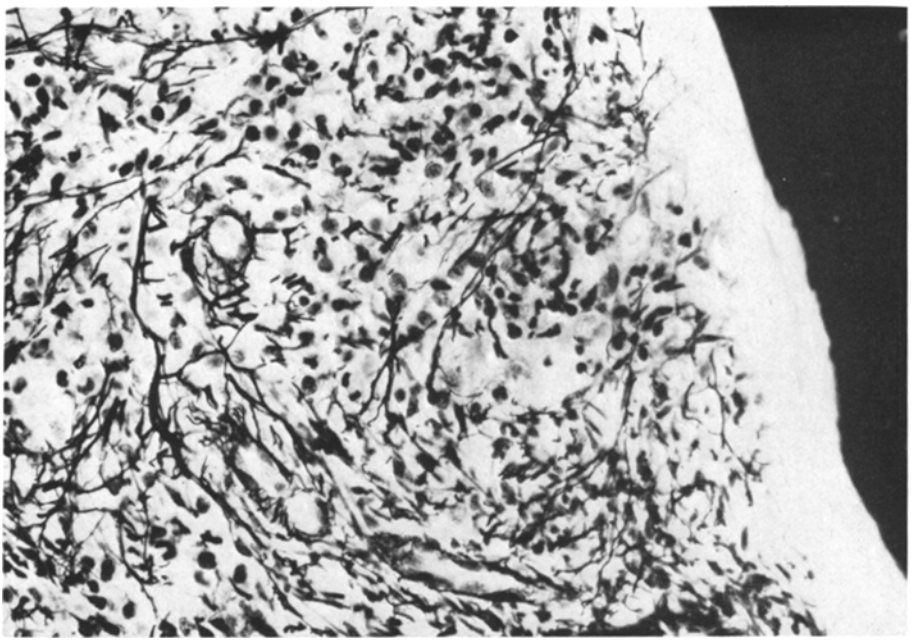

Fig. 2. Iliac crest, needle biopsy $2^1/_2$ months before death. The bone marrow is rich in myelopoietic cells and fibers. Case 2. Gomori, ×350

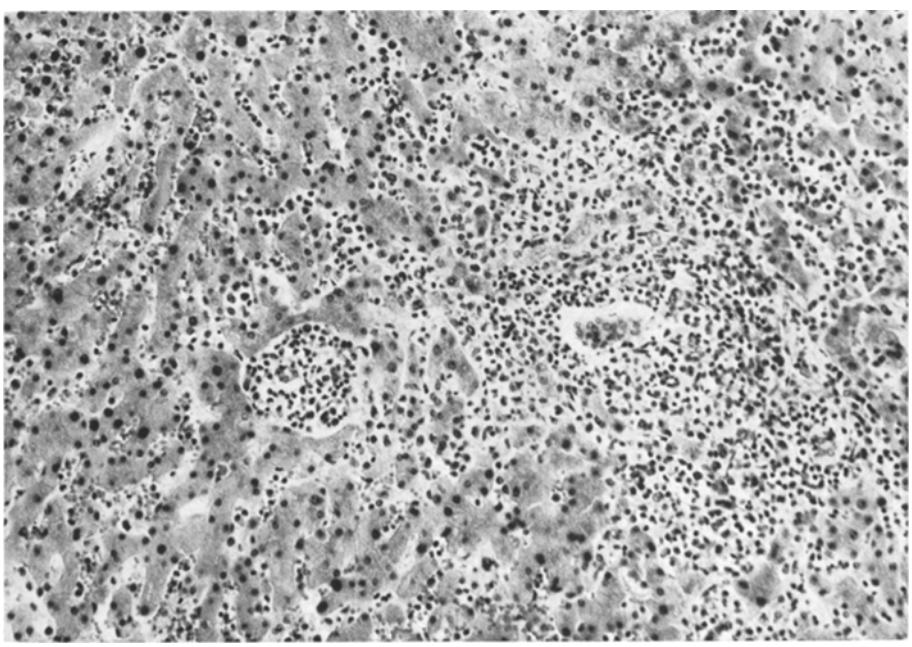

Fig. 3. Liver. Leukemic infiltrates predominantly in the periportal spaces, rich in mature myelopoietic cells. Case 2. H & E, ×140

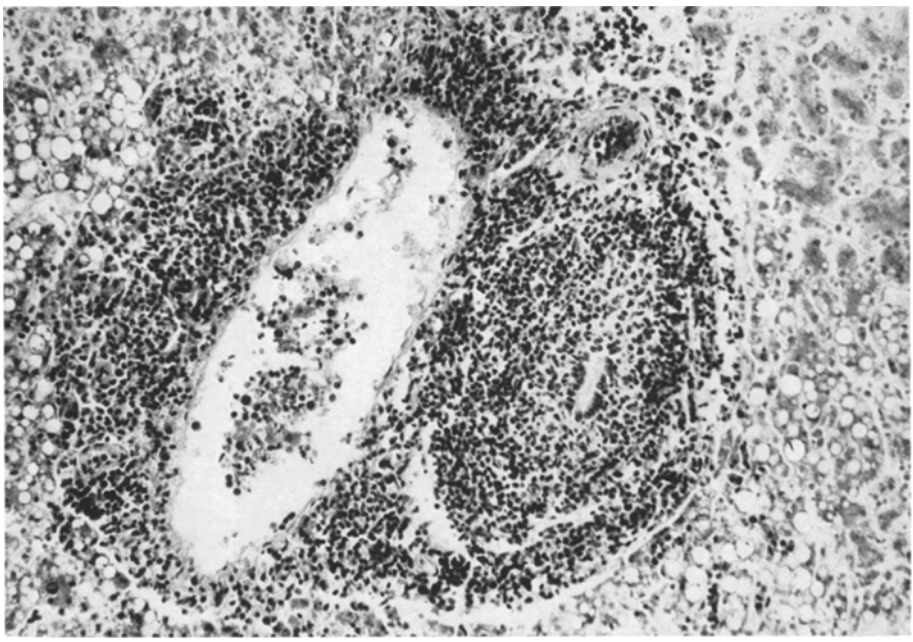

Fig. 4. Liver. Leukemic infiltration of the periportal space. Terminal blast crisis in a case of initially mixed immature-mature myeloid leukemia. 53 year old man (Case 3). H & E, ×140

However, in all but one case particularly peripheral parts of the liver acini had collapsed.

3. Spleen. Three patients had moderate splenomegaly, one massive splenomegaly (the weights are presented in Table 1: 720 g, 400 g, 2125 g, 700 g). We found several up to 3 cm ⌀ large anemic infarcts. The red pulp was diffusely infiltrated with leukemic cells, the trabeculae were dissociated and also infiltrated. The white pulp in Cases 1 and 4 was preserved in large cell-rich foci, whereas in Cases 2 and 3 it was highly diminished. Here, the typical architecture of the spleen was destroyed. In the sinuses there were some macroblasts in Case 3 and some clusters of erythropoietic cells in Cases 2 and 4. On the whole, there were only few megakaryocytes in Cases 2 and 4.

4. Lymph Nodes. There was generalized enlargement (up to 3 cm in diameter) of the lymph nodes in all cases. In Case 2 there was a mediastinal conglomerate of lymph nodes. The tonsils were enlarged and infiltrated with leukemic cells in Cases 3 and 4. The medullary and paracortical pulp was moderately to highly infiltrated with nearly complete destruction of the lymph node architecture in Case 2 (Fig. 5). The capsule, trabecula, and paranodular tissue were locally or diffusely infiltrated. In the sinuses we found erythropoietic cells in Cases 1 and 2, and various amounts of megakaryocytes in Cases 1, 2 and 3. On the whole, there was a slight to moderate increase and in only one case a great increase in the number of reticular fibers and capillaries. Case 3 was complicated by a miliary tuberculosis of the lungs and lymph nodes.

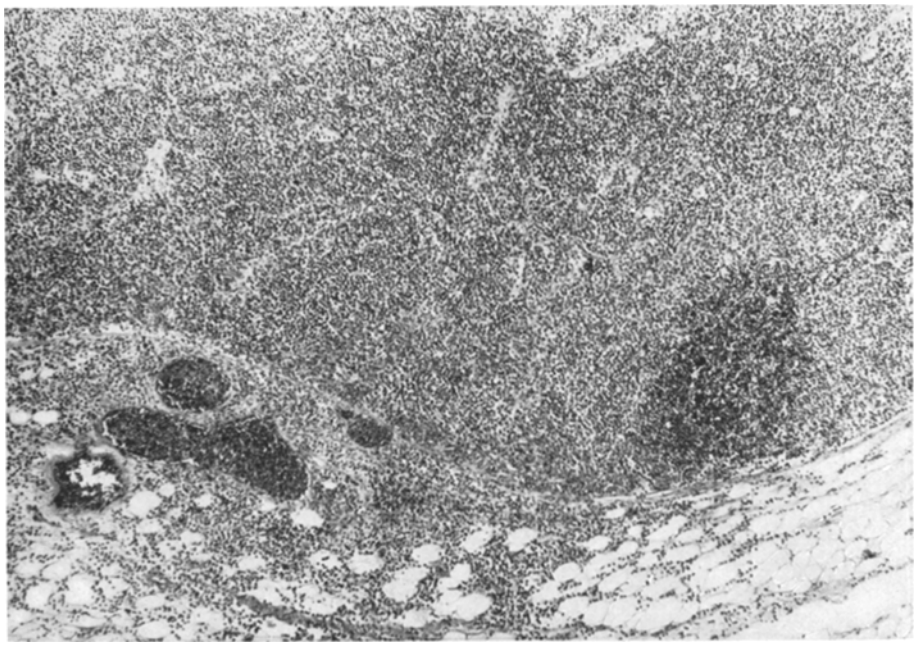

Fig. 5. Lymph node. Great diffuse leukemic infiltration of the paracortical area, cortex, and paranodular tissue. Lymphatic parenchyma only in one focus in the cortex. Predominantly mature myeloid cell picture. Case 2. H & E, ×56

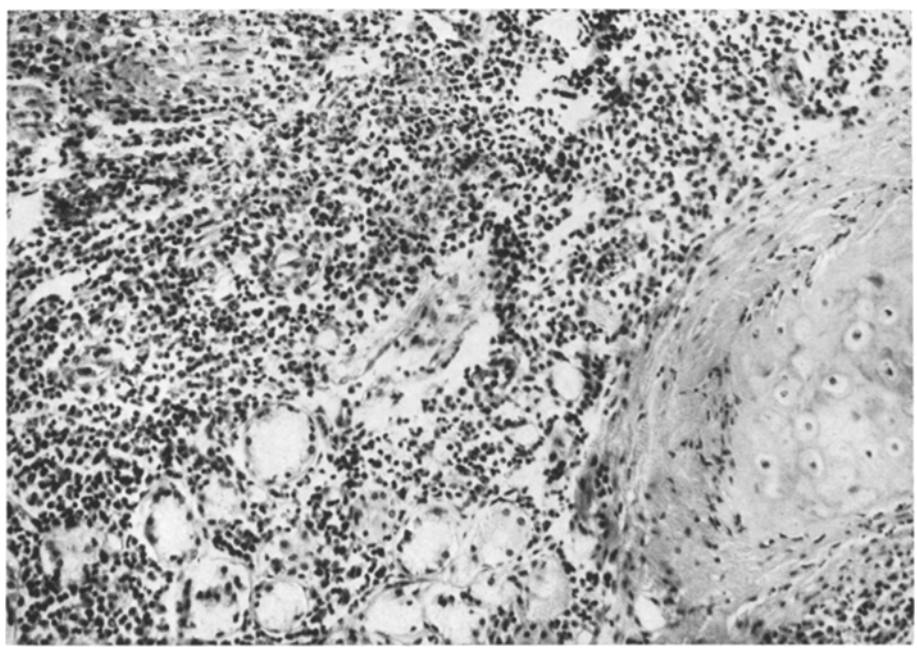

Fig. 6. Lung. Mixed immature and mature myeloid infiltration of the peribronchium. 67 year old man (Case 4). H & E, ×140

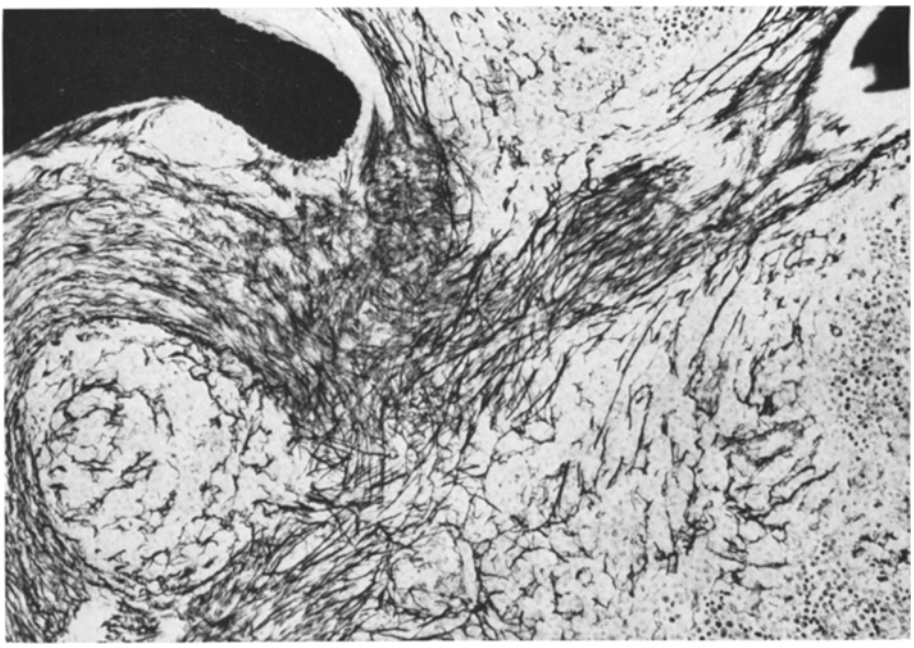

Fig. 7. Vertebra. Peritrabecular and intertrabecular fibrosis. Strands and curls of fibers. Case 4. Gomori, ×140

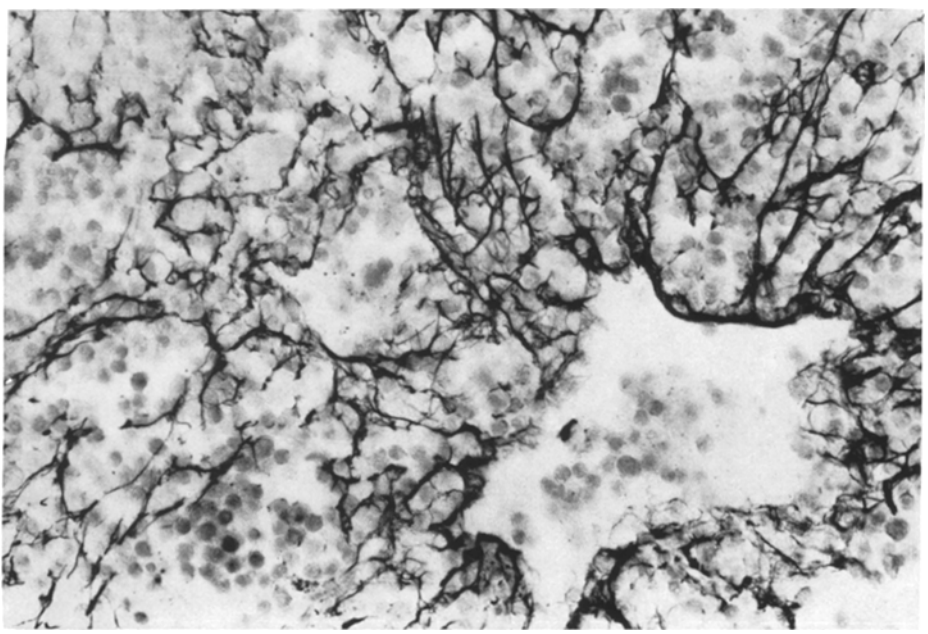

Fig. 8. Vertebra. Fairly dense alveolar fiber network. Moderate ectasia and sclerosis of the sinus. Case 2. Gomori, ×350

5. Lungs. Partly massive leukemic infiltration was to be found in Cases 2 and 4 in the bronchial walls and peribronchium (Fig. 6) and in Case 2 in the pleura and the subpleural lung. We found tumor formation by leukemic infiltrates in the mucosa of bronchi in Case 3.

6. Kidneys. There were small focal leukemic infiltrates in the renal cortex of Case 1. Besides local, sometimes patchy, sometimes striated infiltrates there was an accentuated or diffuse leukemic infiltration particularly in the corticomedullary border region in Cases 2, 3 and 4.

7. Gastrointestinal Tract. A massive leukemic infiltration of the mucosa of the stomach, small bowel, and colon, rich in blast cells, could be seen in Case 3. The intestinal tract of Cases 2 and 4 showed only small leukemic foci.

8. Other Organs. Predominantly small leukemic infiltrates were found in various other organs, such as the cerebrum, the capsule of the hypophysis, and thyroid (Case 2), the adrenal glands (Cases 2 and 4), the testes (Cases 2 and 3), the epididymis (Case 3), the prostate (Case 2), the cutis (Case 3), the gingiva (Case 2), and submandibular glands (Cases 2, 3 and 4).

Discussion

The cases described above show clinical and patho-anatomical similarities characterizing a type of leukemia which differs from typical CML. One of these features was the absence of the Philadelphia chromosome. Others were: no leukocyte counts above 100000 cells/μl, no eosinophilia or basophilia, no increase in the number of thrombocytes, no regular reduction in the reactivity of leukocytes for alkaline phosphatase, no therapeutic effect of busulfan, and a comparatively short life expectancy (Krauss *et al.*, 1964; Lit.; Remy, 1966, Lit.; Hellriegel, 1968; Teplitz, 1968; Mauer, 1969; Ezdinli *et al.*, 1970; Grozdea *et al.*, 1970; Drescher and Simon, 1973). One of the characteristic morphological differences between our Ph^{1-} CML and Ph^{1+} CML is the distribution of the leukemic infiltrates in the liver and other organs. In the liver there was a pronounced leukemic infiltration of the periportal spaces in Ph^{1-} CML, whereas typical CML prefers the sinusoids and infiltrates the periportal spaces as a rule only in the blastic crisis. Leukemic infiltrates of Ph^{1-} CML are also to be found in tissues and organs which are not as frequently or as strongly infiltrated in Ph^{1+} CML, except when the latter terminates in a blastic crisis. These organs include in particular the lymph nodes and the lungs. Furthermore, in two of our 4 cases the white pulp of the spleen was preserved in relatively large foci, although the red pulp was strongly infiltrated with leukemic cells.

The pattern of leukemic infiltration in the liver and spleen, realized in two cases, corresponded to that of the juvenile CML of childhood (Smith and Johnson, 1974), which is often Ph^1 negative. Therefore, the Ph^{1-} type of CML has also been called the juvenile type of CML in adults (Bauke and Bach, 1972; Altman *et al.*, 1974).

Enlargement of the liver and spleen is nearly always a symptom of Ph^{1+} CML. Splenomegaly is even highly typical. However, in Ph^{1-} CML hepatosplenomegaly is less frequent (Ezdinli *et al.*, 1970) and of a lower degree (Remy, 1966). Only one of our cases showed an excessive splenomegaly, and this was the

case with a terminal increase in the number of blasts and with the greatest infiltration of the lungs.

In Ph^{1-} CML the lymph nodes can be strongly infiltrated and therefore enlarged at an early stage. In Ph^{1+} CML, on the other hand, enlargement of the lymph nodes is not observed, except in a blastic crisis or when there is a second Ph^1 chromosome. Duvall *et al.* (1967) observed great enlargement of the lymph nodes in CML with double Ph^{1+} and spoke of CML with lymphoma. In our cases the leukemic infiltration of the lymph nodes was not restricted to the areas typical of Ph^{1+} CML—the hilar pulp and foci in capsule and trabeculae—but was to be found throughout the medulla, paracortical area, and cortex. This pattern corresponds to that of an acute myeloid or myelomonocytic leukemia. This was also true for the leukemic infiltration of the kidneys in our Cases 2, 3 and 4.

The bone marrow and the extramedullary infiltrates of Ph^{1-} CML in children show a wide range of maturation in the myelopoietic series, which differs from case to case (Takanashi, 1972). This is also true for Ph^{1-} CML in adults, according to the clinical observations of Krauss *et al.* (1964) and Hellriegel (1968) as well as to our own autopsy findings. The blood smears, bone marrow, and leukemic infiltrates of our Case 1 shows that it was of a highly mature cell type, comparable to the neutrophilic leukemia of Rubin (1966). In contrast, Case 3 was rich in blasts. Cases 2 and 4 were between these extremes. The large percentage of monocytes in Case 4 shows an association with myelomonocytic leukemia, which has been demonstrated for Ph^{1-} CML in adults (Nowell and Hungerford, 1962) and which seems to be proven for the Ph^{1-} CML of childhood (Altman *et al.*, 1974).

In contrast to the cases of juvenile CML in childhood observed by Takanashi (1972) and Smith and Johnson (1974), in our cases the number of intramedullary erythropoietic cells was greatly reduced. There was also myelofibrosis of at least a low grade. In a synopsis of our findings, Cases 2 and 3 with moderate to severe myelofibrosis were not special forms of acute idiopathic myelofibrosis. The number of megakaryocytes was greatly reduced in two cases, but quantitatively and qualitatively inconspicuous in the two other cases. In all our cases the extramedullary erythro- and megakaryocytopoiesis were much weaker than the myelocytic leukemic infiltration.

The myelofibrosis in myeloid leukemia of the mature cell type is a prognostically unfavorable sign-in Ph^{1-} CML with a short life expectancy as well as in Ph^{1+} CML (Gralnick *et al.*, 1971; Theologides, 1972). Myelofibrosis in Ph^{1+} CML initiates the preterminal phase of the disease (Gralnick and Bennett, 1970).

Whang-Peng *et al.* (1968) assumed that Ph^{1+} CML and Ph^{1-} CML have a different etiology and are different diseases. They drew these inferences from the different therapeutic susceptibility and the different clinical course. However, a greater study of autopsy material will have to show whether our findings in four cases of Ph^{1-} CML are typical, if not specific for this type of leukemia.

The clinical differential diagnosis between Ph^{1+} CML and Ph^{1-} CML is possible only by using a proper technique for the preparation of chromosomes and by following the development of the disease. In any individual the distinction is also not possible on the basis of a needle biopsy of the bone marrow (Whang-Peng *et al.*, 1968). The sometimes pronounced myelofibrosis gives rise to other differential diagnostic questions. However, myelofibrosis alone is not diagnostic

for any form of myeloproliferative disease (Lennert *et al.*, 1975). Clinical features which indicate a Ph1– CML at an early stage will be described by Klein *et al.* (1975).

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