

VOL. 53, NO. 4

APRIL 1979

BLOOD

*The Journal of
The American Society of
Hematology*

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GRUNE & STRATTON

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EXHIBIT

APRIL 1979

Chromosome Analysis in Two Unusual Malignant Blood Disorders Presumably Induced by Benzene

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Two patients with presumably benzene-induced malignant blood disorders with preleukemic phases were cytogenetically monitored through the courses of their diseases. Patient 1, in addition to a familial chromosome translocation [t(3;16)], developed karyotypic abnormalities in 100% of the marrow cells, including two transloca-

tions: t(9;10) and t(4;15). Monosomy of chromosome 7 characterized the cells of patient 2. Cytogenetic monitoring of the patients at various phases of their diseases served as an important indicator of the transformation or progression of the preleukemia into frank leukemia and of the unusual behavior of such leukemic cells.

IT IS GENERALLY ACCEPTED that chronic exposure to benzene (benzol) may induce leukemia in man. Early reports on benzene leukemia in the nineteenth century were followed by an abundant literature on the subject. On the basis of the clinical and hematologic picture, so-called preleukemic phases have been delineated, of which some are nonproliferative (e.g., leukemoid reactions).

Chromosome studies have concentrated mainly on the clastogenic effects (i.e., chromosome breaks and other structural changes) of benzene on lymphocytes in vivo and on a few cases of leukemia (without banding). The exact significance of the chromosome aberrations induced in lymphocytes or bone marrow cells by benzene (or other environmental agents) is not well understood, and the cytogenetic abnormalities reported in benzene leukemia seem to indicate that these changes may not be different from those found in other acute myeloproliferative or lymphoproliferative disorders.¹

This report deals with 2 patients in whom presumably benzene-induced malignant conditions with preleukemic phases were cytogenetically monitored from shortly after the onset of symptoms until death. Some unexpected and unusual dynamics of the malignant cells were revealed.

MATERIALS AND METHODS

Patient 1

Patient 1, a 25-yr old draftsman on steel plates for about 4 yr, used petrol to clean the metal sheets. The solvent, intended for use only as a motor car fuel, contained 2.2% benzene, 7.6% toluene, and 8.7% xylene. It was used in a small unventilated room, and inhalation of the petrol vapor was sufficient to cause nausea. His past history was negative.

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Submitted July 25, 1978; accepted November 17, 1978.

Supported in part by a grant from the ASLK Cancer Fund, Belgium. G. David is an Aspirant of NFWO-België.

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First episode. In September, 1975, the patient complained of slight pyrexia, tiredness, a bleeding tendency, and diffuse arthralgia. He was treated for a few days with an antibiotic for a sore throat and improved when treated with triamcinolone (4 mg/day).

When first seen by us (September 18, 1975), all therapy was withdrawn. He showed only a few ecchymoses; and the spleen was palpable under the costal margin. The blood picture (Table 1A) was mainly characterized by marked leukocytosis (up to 110,000/cu mm) and a marked shift to the left. The granulocytes showed marked toxic granulation, with a normal leukocyte alkaline phosphatase (LAP), which became supranormal during the further evolution of the condition. Plasma acid levels and lactic dehydrogenase activity were markedly increased. All other biochemical data in blood and urine were normal. A slight increase in serum immunoglobulin levels was noted.

The leukocyte count decreased spontaneously during the following weeks; in fact, leukopenia developed. Anemia appeared during the first month of observation, but the thrombocyte count remained normal during that time. Bone marrow (September 24, 1975) showed marked granulocyte hyperplasia with a left shift and marked toxic granulation (Table 1B). Erythroblasts and megakaryocytes were well represented. Within 1 wk the bone marrow became hypocellular, although the erythroblasts were still present in significant numbers. During October and November, 1975, progressive normalization of peripheral blood and bone marrow occurred. A left shift in the peripheral blood and a slight increase of

Table 1A. Patient 1: Hematologic Investigation: Blood

	1975				1976			
	19/9	2/10	24/10	4/12	8/1	29/14	11/5	12/5
Hb (g/dl)	13	8.8	13.5	16	17.3	10	8.2	7
Leukocytes ($\times 10^9$ /liter)	110	29	6.9	12	13	23	154	137
Myeloblasts	—	—	—	—	—	15	53	64
Myelocytes	62	8	1	—	6	9	13	6
Nonsegmented neutrophils	24	36	54	76	66	30	15	16
Eosinophils	—	6	7	4	6	29	4	6
Basophils	—	—	—	—	—	3	—	—
Lymphocytes	—	—	—	—	—	—	—	—
monocytes	14	50	38	20	22	15	20	8
Reticulocytosis (%)	—	2.9	3.4	0.6	—	1.5	1.4	1.3
Thrombocytes ($\times 10^9$ /liter)	225	140	150	80	170	190	—	18
NAP score	102	—	—	214	152	—	—	—
LDH (units/liter)	927	182	—	—	—	—	825	929
Uric acid (mg/dl)	11.3	5.9	—	—	—	10.6	11	10.3

Table 1B. Hematologic Investigation: Bone Marrow

	1975					1976	
	24/9	7/10	15/10	22/10	5/12	29/4	6/5
Myeloblasts	11	—	2	1	1	15	80
Promyelocytes N/E	28/-	2/-	14/-	6/-	4/-	3/1	-/1
Myelocytes N/E	44/-	16/1	14/1	16/2	7/-	7/22	-/2
Metamyelocytes N/E	8/-	10/-	7/-	10/2	2/2	2/7	-/2
Bands N/E	2/1	9/2	3/-	16/1	14/3	2/9	-/2
Segmented neutrophils N/E	5/-	10/3	10/2	25/2	22/6	2/9	-/1
Lymphocytes-monocytes	1	53	47	26	39	4	12
Granulocytes/erythroblasts	7.6	0.9	1	1.9	6	24	24
Megakaryocytes	nl.	1	1	nl. 1	1	nl.	abs.
Cellularity	xxx	1	1	nl.	nl.	++	+4

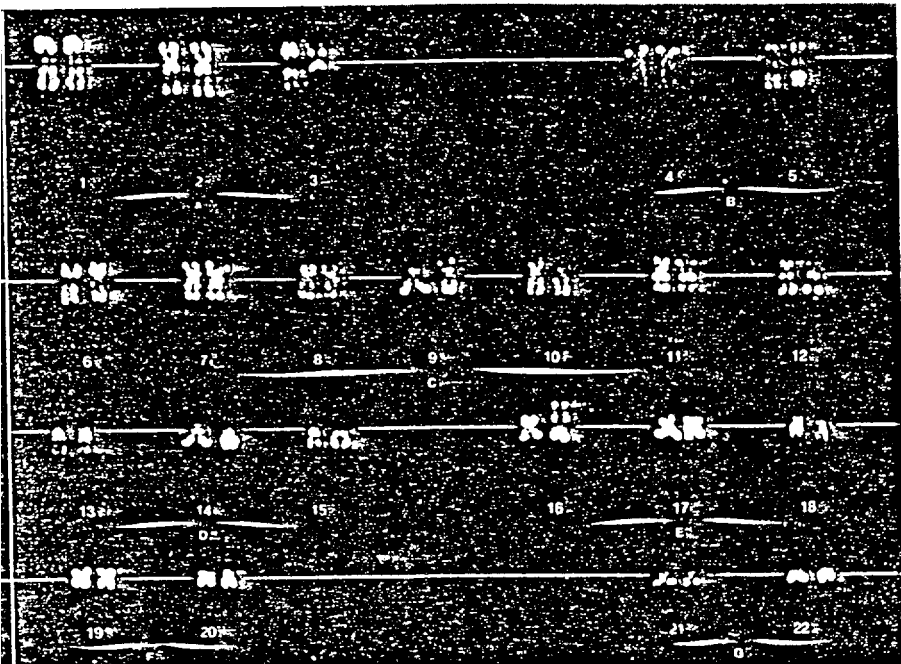


Fig. 1. R-banded karyotype of patient 1 showing the reciprocal translocation $t(9;10)$ in a marrow cell.

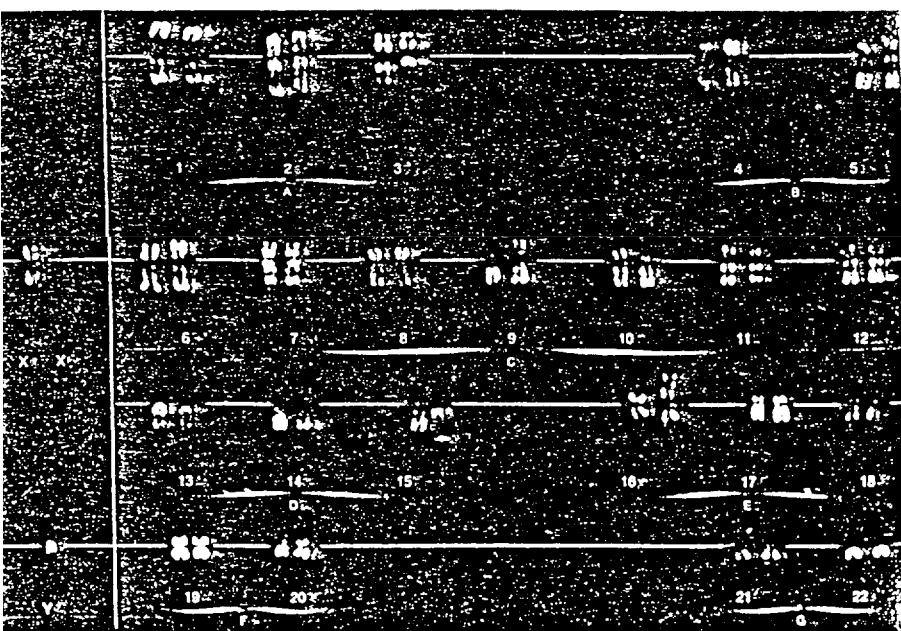


Fig. 2. R-banded karyotype of patient 1 of a cell examined in February 1980. The cell now also contains a $4x2$ in addition to the reciprocal translocation $t(9;10)$.

histiocytes in marrow, liver, and spleen and the diagnosis of CMML was finally confirmed by the presence of the characteristic granules.

From October 1975, until March 1976, no treatment was administered. The patient returned to his work but was unable to lose weight.

Second episode. During the first episode, the patient was treated at home with antibiotics and lost weight. When seen on April 2, 1976, the spleen was palpable 6 cm with a marked left shift and his condition improved slightly, and the hemoglobin and 100,000/ μ m. Anemia and LDH activity increased markedly. T eosinophils, and erythroblastopenia. C blasts, and the only mature cells were diagnosed as acute myelomonocytic leukemia. Therapy with cytosine arabinoside was started on May 16, 1976. During this second

Table 2A. Patient

Hb (g/dl)
Leukocytes ($\times 10^9$ /liter)
Myeloblasts
Myelocytes
Nonsegmented neutrophils
Eosinophils
Basophils
Lymphocytes
Monocytes
Reticulocytosis (%)
Thrombocytes ($\times 10^3$ /liter)
LDH (units/liter)
Uric acid (mg/dl)
NAP score

Table 2B. Patient

Myeloblasts
Promyelocytes N/E
Myelocytes N/E
Metamyelocytes N/E
Bands N/E
Segmented neutrophils N/E
Lymphocytes
Monocytes
Plasmocytes
Granulocytes/erythroblasts
Megakaryocytes
Cellularity

eosinophils in peripheral blood and bone marrow were noted. Slight reticulocytosis was present. A diagnosis of CML was initially considered but rejected because of the absence of a Ph^1 chromosome, the presence of the marked toxic granulation in granulocytes, and the normal or increased LAP scores.

From October, 1975, until March, 1976, the clinical state was normal; the spleen was not palpable. No treatment was administered from September 18, 1975, to April 1, 1976. During this period the patient returned to his work but was no longer exposed to benzene or other organic solvents.

Second episode. During the first week of April, 1976, the patient developed a pulmonary infection and was treated at home with antibiotics. His condition deteriorated; he had pyrexia up to 39.8°C , and he lost weight. When seen on April 22, 1976, the patient appeared acutely ill. He had a productive cough, and the spleen was palpable 6 cm under the costal margin. The leukocyte count was $93,000/\text{cu mm}$, with a marked left shift and eosinophilia of 39%. After treatment with antibiotics and allopurinol, his condition improved slightly, and the leukocyte count showed marked oscillations between $23,000/\text{cu mm}$ and $100,000/\text{cu mm}$. Anemia and thrombocytopenia developed, and plasma uric acid levels and LDH activity increased markedly. The bone marrow on April 29, 1976, showed 15% blasts, 47% eosinophils, and erythroblastopenia. On May 6, 1976, 80% of the bone marrow cells were myelomonoblasts, and the only mature cells were eosinophils. Erythroblasts and megakaryocytes were absent. The diagnosis of acute myelomonocytic leukemia was established, and treatment with thioguanine and cytosine arabinoside was started on May 9, 1976, together with antibiotics and intensive supportive therapy. Septicemia due to enterobacter and *Pseudomonas* developed. The treatment was without effect: the cardiopulmonary condition worsened, the spleen increased further in size, and the patient died on May 16, 1976. During this second period, no further biochemical anomalies developed. At autopsy,

Table 2A. Patient 2: Hematologic Investigation: Blood

	4/8/76	3/9/76	10/9/76	20/9/76	22/9/76
Hb (g/dl)	9.3	6	8	9.4	9
Leukocytes ($\times 10^9/\text{liter}$)	29.8	1.2	1.1	2.0	2.3
Myeloblasts	—	—	—	—	—
Myelocytes	16	—	—	—	—
Nonsegmented neutrophils	43	18	—	66	79
Eosinophils	1	—	—	—	—
Basophils	—	—	—	—	—
Lymphocytes	25	80	100	31	17
Monocytes	16	2	—	3	4
Reticulocytosis (%)	0.1	—	—	—	—
Thrombocytes ($\times 10^9/\text{liter}$)	—	20	4	8	28
LDH (units/liter)	490	—	—	134	—
Uric acid (mg/dl)	9.3	—	—	—	—
NAP score	210	—	—	—	—

Table 2B. Patient 2: Hematologic Investigation: Bone Marrow

	14/8/76	23/8/76	4/9/76	13/9/76	20/9/76
Myeloblasts	7	—	—	—	2
Promyelocytes N/E	12/-	—	2/-	1/-	9/-
Myelocytes N/E	13/-	1/2	4/-	—	6/-
Metamyelocytes N/E	20/-	2/1	1/-	—	10/-
Bands N/E	20/-	13/2	2/2	—	6/-
Segmented neutrophils N/E	17/1	12/4	25/3	1/-	9/-
Lymphocytes	8	15	53	75	6
Monocytes	2	38	—	—	1
Plasmacytes	—	—	7	23	41
Granulocytes/erythroblasts	—	—	—	—	3.9
Megakaryocytes	abs.	+	abs.	abs.	+
Cellularity	+++	±	11	11	++

pulmonary infiltration, diffuse edema, hemorrhages, and marked splenomegaly (4000 g) were found. Infarctions were found. Leukemic proliferation was present in most organs.

Patient 2

Patient 2, a 67-yr-old retired carpenter, suffered from gastric ulcers during 1953–1957 during which time his wife died from leukemia. He also had recurrent episodes of depression. During his professional life as a carpenter he had been using glues containing benzene for many years. Beginning in May, 1976, he complained of tiredness, nocturnal sweats, and loss of appetite. Medical examination at that time revealed anemia and bone marrow compatible with megaloblastic anemia. Vitamin B₁₂ was administered without success.

At the end of June, 1976, he was markedly anemic, and an increased leukocyte count (23,400/cu mm) with a marked shift to the left was present (Table 2A). The LAP score was increased to 210 (July 1976) (normal 14–100).

Plasma uric acid was 8.4 mg/dl, iron was 199 µg/dl, and TIBC was 444 µg/dl. All other serum chemistries were normal. The bone marrow showed increased cellularity due to myeloid hyperplasia with only occasional erythroblasts and megakaryocytes. All myeloid maturation stages were present; however, degranulation was marked, and Dohle bodies were present. Monocytosis (Table 2B) was observed on one occasion (August 13, 1976).

Because of the high LAP score and the absence of a Ph¹ chromosome, CML was rejected, and a provisional diagnosis of a leukemoid reaction was made. Extensive investigations, including lymph node biopsy, during the following months failed to reveal any infection or malignancy. Pending the results of the Lowenstein culture (it was later known that all remained negative for acid-fast bacteria), a 1-month trial with tuberculostatic drugs was inaugurated, without clinical benefit. The patient came under home care early in August, 1976. His condition was essentially unchanged since June, 1976. The anemia was marked, and repeated transfusions were necessary. *In the absence of any cytostatic or antibiotic treatment*, the leukocyte and thrombocyte counts dropped progressively. On August 23, hypocellular bone marrow was found, and after September 4 all bone marrow samples were entirely aplastic.

The serum LDH level reverted to normal, and it was thought that the earlier increase in LDH was due to inefficient proliferation of the bone marrow.

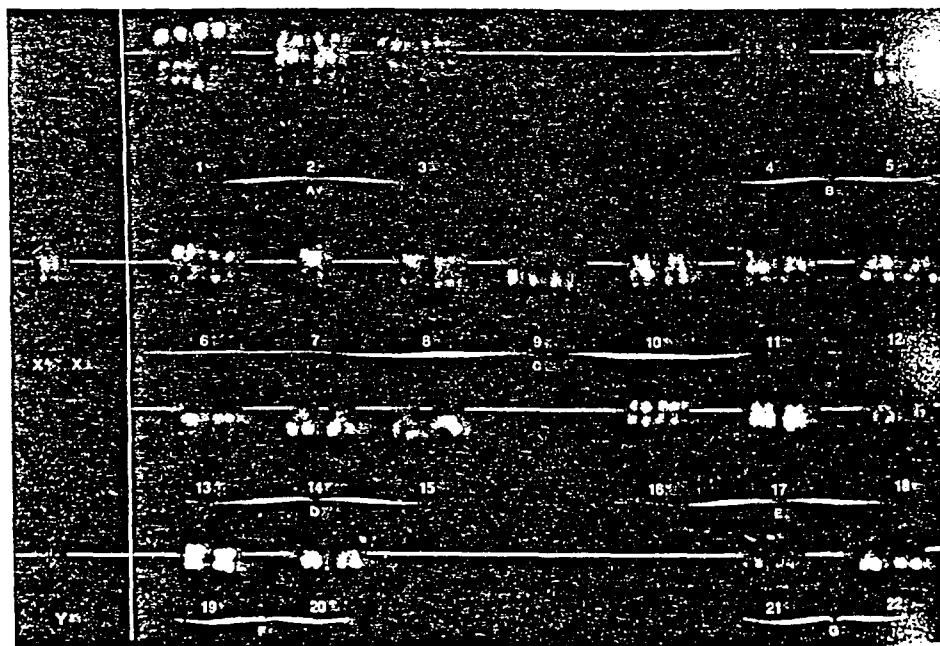


Fig. 3. A-banded karyotype of a marrow cell of patient 2 showing the monosomy of chromosome 7.

The clinical course was mainly characterized by an insidious onset with cytopenia. Nevertheless, the patient had (Table 1) a just bone marrow examination with granulocytic hyperplasia. Numerous mature plasma cells were present.

At autopsy, three recent gastric ulcers were present. Extensive pulmonary infection and necrosis were present. The bone marrow showed slight extramedullary hematopoiesis.

Cytogenetic Investigations

Chromosome preparations were made and were processed for Banding. The presence of PHA for 72 hours to get was evaluated by screening of 10 cells shown in Table 3.

Patient 1 had a familial translocation 3 and the short arm of chromosome 3 present in the patient's lymphocytes. The bone marrow and

Table 3

Date	M/B = PHA	444
Patient 1		
24 Sep 75	M	2
24 Sep 75	B - PHA	2
6 Oct 75	B - PHA	—
6 Oct 75	B - PHA	—
6 Oct 75	M	—
22 Oct 75	M	—
30 Oct 75	M	—
20 Nov 75	M	2
18 Dec 75	M	3
29 Jan 76	M	2
29 Apr 76	M	1
29 Apr 76	B - PHA	—
8 May 76	M	—
7 May 76	B - PHA	—
Patient 2		
1 Jul 76	M	6
1 Jul 76	B - PHA	—
12 Aug 76	M	10
12 Aug 76	B - PHA	2
23 Aug 76	M	—
23 Aug 76	B - PHA	—
1 Sep 76	B - PHA	—
3 Sep 76	M	—
13 Sep 76	B - PHA	—
20 Sep 76	B - PHA	—
20 Sep 76	M	1

*Normal and abnormal cells in patient 1.

†Except one cell, which was 46,XY,FT (5).

The clinical course was mainly influenced by a variety of infections and hemorrhagic complications requiring intensive treatment with antibiotics and transfusions of packed erythrocytes and thrombocytes. Nevertheless, the patient's condition deteriorated progressively, and he died from GI hemorrhages (Table 1). A last bone marrow examination on September 20, 1976, showed normal cellularity, toxic granulations in the myeloid cells, and an appreciable quantity of erythroblasts and megakaryocytes. Numerous mature plasmocytes were observed for the first time.

At autopsy, three recent gastric ulcers proved to be the direct cause of the GI hemorrhages and death. Extensive pulmonary infection due to *Aspergillus* and Gram-negative bacteria and marked arteriosclerosis were present. The bone marrow showed normal cellularity, and the atrophic spleen (87 g) showed slight extramedullary hematopoiesis and siderosis.

Cytogenetic Investigations

Chromosome preparations were made from bone marrow and blood cultures of approximately 48 hr and were processed for R banding (acridine orange fluorescence). Blood cells were also grown in the presence of PHA for 72 hr to determine the karyotype in the normal lymphocytes. Y-chromosome loss was evaluated by screening of 100 metaphases with Q banding. The results of the investigations are shown in Table 3.

Patient 1 had a familial translocation (FT) characterized by exchange of the long arm of chromosome 3 and the short arm of chromosome 16 [t(3;16)(q11;p11)]. This chromosome anomaly was found to be present in the patient's lymphocytes and skin fibroblasts as well as in the cells of several members of his family. The bone marrow and peripheral blood cells grown without PHA at the first investigation

Table 3. Summary of the Cytogenetic Investigations

Date	M/B ± PHA	Chromosome Count					Number of Normal Cells*	Number of Abnormal Cells*	Karyotype (R Banding)
		≤44	45	46	47	≥48			
Patient 1									
24 Sep 75	M	2	1	7	—	—	0	10	46,XY,FT,t(9;10)(p24;p12)
24 — 75	B — PHA	2	3	5	—	—	0	10	46,XY,FT,t(9;10)(p24;p12)
6 Oct 75	B + PHA	—	2	23	—	—	24	0	46,XY,FT,normal
6 Oct 75	B — PHA	No mitoses							
6 Oct 75	M	No mitoses							
22 Oct 75	M	—	—	10	—	—	0	10	46,XY,FT,t(9;10)(p24;p12)
30 Oct 75	M	—	2	a	—	—	0	10	46,XY,FT,t(9;10)(p24;p12)
20 Nov 75	M	2	1	20	—	—	6	17	46,XY,FT,normal/46,XY,FT,t(9;10)(p24;p12)
18 Dec 75	M	3	1	16	—	—	2	18	46,XY,FT,normal/46,XY,FT,t(9;10)(p24;p12)
29 Jan 76	M	2	3	13	—	—	1	17	46,XY,FT,normal/46,XY,FT,t(9;10)(p24;p12)
29 Apr 76	M	1	2	12	1	—	0	16	46,XY,FT,t(9;10)(p24;p12),t(4;15)(p13;q14)
29 Apr 76	B — PHA	—	1	14	1	—	0	16	46,XY,FT,t(9;10)(p24;p12),t(4;15)(p13;q14)†
6 May 76	M	—	4	11	—	—	0	15	46,XY,FT,t(9;10)(p24;p12),t(4;15)(p13;q14)
7 May 76	B — PHA	—	2	16	2	—	0	20	46,XY,FT,t(9;10)(p24;p12),t(4;15)(p13;q14)
Patient 2									
1 Jul 76	M	6	8	1	—	—	1	14	45,XY,-7
1 Jul 76	B — PHA	—	4	—	—	—	0	4	45,XY,-7
12 Aug 76	M	10	4	—	—	—	0	14	45,XY,-7
12 Aug 76	B — PHA	2	5	—	—	—	2	5	46,XY,normal/45,XY,-7
23 Aug 76	M	No mitoses							
23 Aug 76	B — PHA	No mitoses							
1 Sep 76	B — PHA	No mitoses							
3 Sep 76	M	—	1	3	—	—	4	0	46,XY,normal
13 Sep 76	B — PHA	No mitoses							
20 Sep 76	B — PHA	No mitoses							
20 Sep 76	M	1	2	9	—	—	12	0	46,XY,normal

* Normal and abnormal cells in patient 1 show a familial translocation (FT) t(3;16)(q11;p11).

† Except one cell, which was 46,XY,FT,(9;10)(p24;p12).

showed an acquired translocation in addition to the constitutional one. Part of the short arm of chromosome 10 was deleted and translocated on the short arm of chromosome 9, i.e., $t(9;10)(p24;p12)$. At that time, 11% myeloblasts were present in the marrow, and the WBC was 31,000/cu mm, with many immature elements. Twelve days later, when the bone marrow had become hypocellular without blast excess and the WBC had spontaneously dropped to 2100/cu mm, marrow and blood cultures failed to show any mitoses. On October 22 the cellularity and cytology of the bone marrow normalized, but the 10 metaphases examined all showed the abnormal karyotype. On October 30 the same result was obtained. On November 20, cytogenetically normal cells showing only the FT appeared in the bone marrow (265%) for the first time, but their number decreased again in December 1975, and January, 1976. Further cytogenetic monitoring was then temporarily suspended because the patient was in good condition and had gone back to work. When he returned a few months later and was found to be overtly leukemic, the acquired karyotypic anomaly persisted, and an additional one had developed, consisting of an almost complete exchange between the long arm of chromosome 15 and the short arm of chromosome 4, i.e., $t(4;15)(q13;q14)$. Two cells with 47 chromosomes showed an extra minute marker. Only one cell was found in which the additional translocation was not present.

Patient 2, on July 1, 1976, had chromosome 7 monosomy in the blood and bone marrow, in which only 1 normal cell out of 15 was found. On August 12 the same anomaly was found in 100% of the cells. No mitoses were found in the bone marrow of August 23 or in the blood of August 23 and September 1. At that time the marrow had become aplastic, the WBC was 1200/cu mm, and almost exclusively lymphocytes were present in the blood. Only four metaphases were found in the marrow of September 3. They were completely normal. On September 20, when the cellularity of the marrow had become normal again, all metaphases showed a normal karyotype.

DISCUSSION

It is very likely, although difficult to prove, that a causal relationship existed between the hematologic disorders and the professional activities of both patients. In the first patient it was proved that he had used solvents containing large quantities of benzene. The second patient apparently had been using glues containing benzene for many years. The use of glues containing more than 1% benzene is unlawful in Belgium. The measure, however, has not always been strictly enforced, and two other carpenters are currently being treated for aplastic anemia in our department. We may surmise that the patient had been exposed to benzene before the law was passed and that the possibility of exposure existed even after that.

Both patients initially presented with so-called preleukemia, as manifested by an increased WBC with immature elements in the blood and an elevated number of blasts in the marrow. This preleukemic state, moreover, was characterized by a clonal type of chromosomal anomaly that was present in 100% of the bone marrow and blood metaphases in patient 1 and in 95% of the bone marrow metaphases in patient 2. Clonal proliferations in man, as a rule, are malignant in nature. Exceptions to this rule may be endothelial cell proliferation in atherosclerosis, nocturnal paroxysmal hemoglobinuria (NPH),¹ and some benign gammopathies, particularly in elderly persons.^{4,5} Ataxia telangiectasia appears to be the only exception in which a consistently demonstrable karyotypic change [$t(14;14)(q12;q32)$] is present many years before malignancy develops.⁶ The significance of clonal abnormalities in some patients with polycythemia vera and myelofibrosis is still obscure. On the other hand, there is convincing evidence that many leukemias may be preceded by a preleukemic stage, such as pancytopenia or aplastic anemia, in which consistent chromosomal abnormalities exist in the marrow. Not only is the proportion of these preleukemias presenting chromosomal anomalies with clonal character similar to that found in AL, but during the preleukemia the same karyotypic anomaly that was demonstrated in the preleukemic

phase characterized the leukemia during the leukemic phase. The malignant process in the absence of an antecedent 100,000/cu mm leukocyte count returned to normal and a picture again, with the exception cells began to appear in the abnormal. Hence, the pathologic and cytologic remission that the disease may number of bone marrow relapse occurred.

In patient 2 the course of the disease, however, disappeared total bleeding and infectious complications started to recover and was on two occasions. From the rejection of the presumption of normal hematopoiesis, and of plasmocytosis in rejection may have occurred partial and of short duration whose death from complications.

Spontaneous regression in no cases with abnormal karyotype documented. The chromosomal frequently associated with regularly found in acute leukemia was found in patient 2, is an anomaly in AML; only trisomy 11.

Chromosome studies in leukemia have been reported,⁹⁻¹³ but we have used these techniques. One case of candidate) is mentioned by (one benzene-induced preleukemia). Unfortunately, no banding techniques were used, whether a (9;22) or another translocation, in benzene-induced leukemia. anomalies that occur nonclonally in proliferative disorders would have the significance of these anomalies is unlikely, indeed, that they also appear in benzene leukemia, and questions about the relationship between cancer and specific chromosomal anomalies.

Patient 1 was a carrier of the (9;22) translocation. Whether it

phase characterizes the leukemic cells. Further karyotypic evolution may occur during the leukemic phase.'

The malignant process in our 2 patients showed a remarkable natural history. In the absence of any antitumor treatment, the WBC of patient 1 dropped from over 100,000/cu mm to about 2000/cu mm within 1 wk, the myeloblasts in the marrow returned to normal, and after a hypoplastic phase the marrow showed a normal picture again, with the exception of an increased number of eosinophils. Normal cells began to appear in the marrow, in which initially 100% of the metaphases were abnormal. Hence, the patient spontaneously went into a seemingly complete clinical and cytologic remission, allowing the patient to return to work. The only sign that the disease may not have disappeared totally was that a substantial number of bone marrow metaphases was still abnormal; indeed, 3 mo later a fatal relapse occurred.

In patient 2 the course of the disease was very similar. His aneuploid cell clone, however, disappeared totally, leaving an aplastic marrow. The patient died from GI bleeding and infectious complications at a time when his bone marrow had already started to recover and was accompanied by plasmocytosis and normal metaphases on two occasions. From these observations it can be concluded that spontaneous rejection of the presumably malignant cells occurred, with (temporary) regeneration of normal hematopoiesis. The concurrent appearance of eosinophilia in patient 1, and of plasmocytosis in patient 2, may lead support to the hypothesis that this rejection may have occurred on an immunologic basis. However, rejection was only partial and of short duration in patient 1; it may have been complete in patient 2, whose death from complications prevented further follow-up.

Spontaneous regression of leukemia in man has been repeatedly observed,¹ but no cases with abnormal karyotype, studied with banding techniques, have ever been documented. The chromosomal anomaly found in patient 1 is not known to be frequently associated with any particular malignancy, but such translocations are regularly found in acute leukemia. The chromosome 7 monosomy, however, which was found in patient 2, is the second most frequently encountered chromosome anomaly in AML; only trisomy 8 is more frequent.¹

Chromosome studies in benzene-induced malignant hematologic disorders have been reported,⁹⁻¹³ but we know of no cases that have been studied with banding techniques. One case of monosomy C (statistically monosomy 7 is a strong candidate) is mentioned by Aksoy,¹⁴ who also stressed the finding in two instances (one benzene-induced preleukemia and one benzene-induced leukopenia) of a Ph¹. Unfortunately, no banding or pictorial material was presented, and it is unknown whether a (9;22) or another type of translocation was present. The clear demonstration, in benzene-induced malignancy, of a Ph¹ or other structural chromosome anomalies that occur nonrandomly in human myeloproliferative and lymphoproliferative disorders would be of utmost importance for a better understanding of the of these anomalies in malignant processes. It would seem rather unlikely, indeed, that these characteristic chromosome anomalies, if they should also appear in benzene leukemia, would be of primary importance to leukemogenesis, and questions about the relationship between individual etiologic agents in cancer and specific chromosome anomalies would lose some of their interest.¹⁵⁻¹⁸

Patient 1 was a carrier of an apparently balanced and familial (3;16)(q11;q11) translocation. Whether malignancy occurs more frequently in individuals with

constitutionally abnormal karyotype other than trisomy 21 or occurs in clusters in families in which constitutional chromosome anomalies have occurred remains an open question.¹

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Effect of Antiplatelet

We studied the interaction of human platelet activated by IgG obtained from 3 patient fusion purpura syn heparin-plasma buffer complement activated platelet function. With ment activation was immune alteration of P1^{AI} antibody was presence of complement both platelet lysis and (release reaction) in were observed. Plate by ⁵¹Cr loss, require body than was neces

PLATELET IMMUNITY may be mediated by immune complexes of immune alterations of IgG-coated platelet result.¹⁰⁻¹² Complement lysis of human platelet alteration mediated man.^{14,15}

We studied the conditions in which antibody obtained We studied the natural component require antibodies can mediate platelets.

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Submitted July 7, 1978.
Supported by American Cancer Society and by Special Fellowship of American Leukemia Society of Philadelphia.
Presented in part before the American Society of Hematology, 1978.

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