A New Case of Isolated Tetrasomy of Chromosome 8 in a Patient with Therapy-Related Myelodysplastic Syndrome: Confirmation by Chromosome Painting in Metaphase and Interphase Nuclei

Isolated trisomy 8 is a frequent cytogenetic finding in acute nonlymphocytic leukemia (ANLL) and myelodysplastic syndromes (MDS) [1]. In contrast, isolated tetrasomy 8 has rarely been reported in these disorders [1–5]. We report a case of therapy-related MDS with isolated tetrasomy 8, which was identified by both conventional cytogenetic analysis and chromosome painting, one of the varied fluorescent in situ hybridization (FISH) techniques [6] that allow characterization of numerical and structural chromosome aberrations.

A 46-year-old man was referred in January 1992 for anemia and leukopenia. In 1987, he had been treated for Hodgkin's disease by three courses of MOPP and mantle-field irradiation. On admission, he had no organomegaly. The blood count showed anemia: hemoglobin level 11.2 g/dl, mean corpuscular volume 108 μ^3, leukopenia (leukocytes 3×10^9/L, with 32% neutrophils, 64% lymphocytes, 4% monocytes), and platelet count 115×10^9/L. Eight percent of the circulating nucleated cells were erythroblasts. Bone marrow (BM) aspirate was hypocellular, containing 7% blasts, 45% erythroblasts, and myelodysplastic features involving all myeloid series, with no ringed sideroblasts. BM biopsy was normocellular and showed moderate fibrosis. Therapy-related MDS with myelofibrosis was diagnosed. The patient has very recently been allografted from his HLA identical brother.

BM and peripheral blood (PB) cells without stimulation were analyzed cytogenetically after 24-hour cultures. Five metaphases in the BM and 13 metaphases in the PB showed tetrasomy 8 (Fig. 1). One mitosis in the BM, but none in the PB, was normal.

Classic cytogenetic analysis was completed by a FISH technique. The whole chromosome painting system commercially available (GIBCO-BRL Life Technologies), using whole chromosome 8 nonradioactive probes directly labeled with fluorophore, was used. These probes, made from flow-sorted human chromosomes [7–10], were hybridized on BM and PB metaphase and interphase nuclei. The manufacturer's protocol was applied with slight modifications: slides were postfixed with 3.7% formaldehyde in phosphate buffered saline (PBS) to preserve the chromosome structure and rinsed three times with PBS before denaturation and dehydration in ice-cold ethanol series. Posthybridization washes were performed at 42°C. Spreads were visualized on a microscope equipped with epifluorescence and with an N2 Leitz filter set (rhodamine).

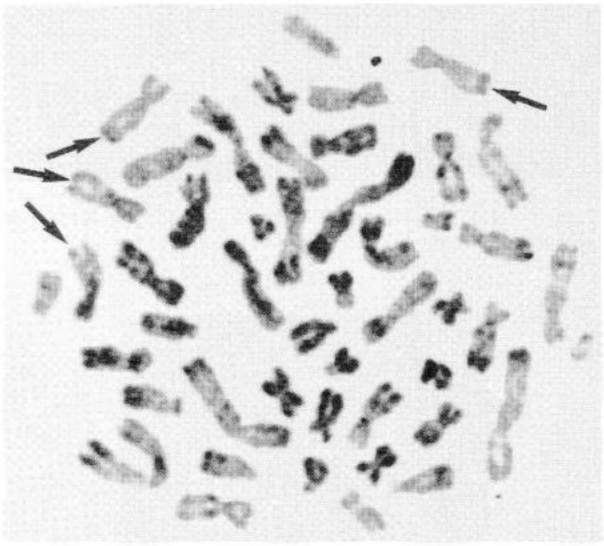

Figure 1 Bone marrow metaphase (R-banded) showing tetrasomy 8 (arrows) in a case of therapy related MDS.

All metaphases scored showed four painted chromosomes (Fig. 2), and most interphase nuclei clearly exhibited four fluorescent domains (Fig. 3). The results completely confirmed those of the standard cytogenetic method.

Although tetrasomy of chromosome 8 has been observed in association with other chromosome anomalies in ANLL and the blastic phase of chronic myelocytic leukemia [1], isolated tetrasomy 8 is a very rare finding, apparently reported only five times [1–5]. Three of those patients had ANLL of M4 or M5 type, and one patient had MDS. The remaining patient had polycythemia vera, treated for 2 years by hydroxyurea, and a minor +8, +8 clone suggesting, as in our patient, a therapy-related occurrence.

In our patient, FISH detected tetrasomy 8 in all metaphase and interphase cells examined and thus proved at least as sensitive as conventional cytogenetic analysis, which had demonstrated tetrasomy 8 in all but one of the mitoses examined. In chronic lymphocytic leukemia, FISH has proved superior to conventional cytogenetics in detecting trisomy 12 [11]. In ANLL and MDS, it will be of interest to determine whether FISH can detect numerical anomalies (especially

Cancer Genet Cytogenet 65:175–176 (1993)
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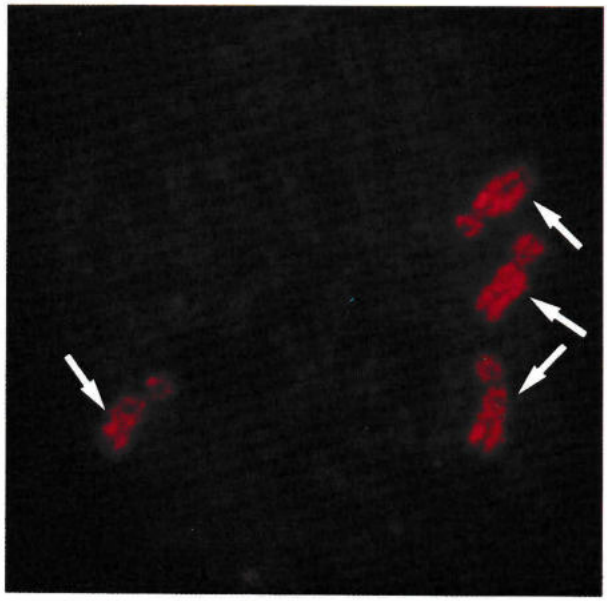

Figure 2 Metaphase spread showing four painted chromosomes 8 (arrows).

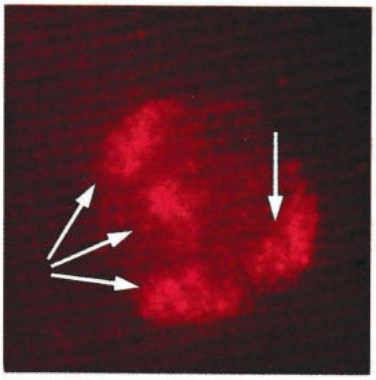

Figure 3 Interphase nuclei displaying four hybridization signals (arrows).

those involving chromosomes 7 and 8) with a higher frequency than conventional cytogenetics. This could stress even further the pathogenetic importance of such abnormalities in MDS and AML and also might allow use of FISH in detection of residual disease after treatment. It will also be of interest to determine whether centromeric repetitive probes will prove superior to whole chromosome probes by avoiding risks of coalescent signals [12].

This work was supported by the Fondation de France contre les Leucémies. Color illustration costs were in part provided by a grant from GIBCO-BRL Life Technologies.

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