

Persisting Clone of Cells With an Abnormal Chromosome in a Woman Previously Irradiated

Anatole Dekaban, M.D.¹

Bethesda, Maryland

During family studies of dystrophic and malformed children, we have encountered persistent chromosome abnormalities in the blood cultures of a 32-year-old mother. She has been in good health except for the recurrent eczematous dermatitis for which she has received repeated x-ray treatment since age 16. Her family history was negative for the presence of congenital malformations, mental retardation or evident hereditary disorders. She had a total of five pregnancies: the first resulted in miscarriage at three months, the next three led to the birth of normal children and the last pregnancy resulted in a severely retarded and malformed male. Chromosomes were processed from three cultures of her blood drawn at six and nine month intervals and also from the skin biopsy. In addition, chromosomes preparations were made from the blood cultures of her abnormal child, her father, and her husband. The chromosomes were scored, and all mitoses with chromosomal abnormalities were photographed and karyotyped. The mother's chromosomes prepared from the blood cultures had two types of the abnormalities, whereas, the chromosomes of her malformed child, her father and her husband were normal.

RESULTS

Summary of the data on x-ray treatment which this woman received:

1947—five irradiations to both ankles

1951—three irradiations to the back of the neck and occipital areas

1956-1957—15 irradiations to various parts of her body

1964—four irradiations to hands and wrists

The following is an example of parameters of radiation used in one of the treatments: 100 KV, 5 ma, total filter of aluminum 0.6 mm, distance 30 cm, rate

¹Section on Child Neurology, SN, NINDB, National Institutes of Health, Bethesda, Maryland.

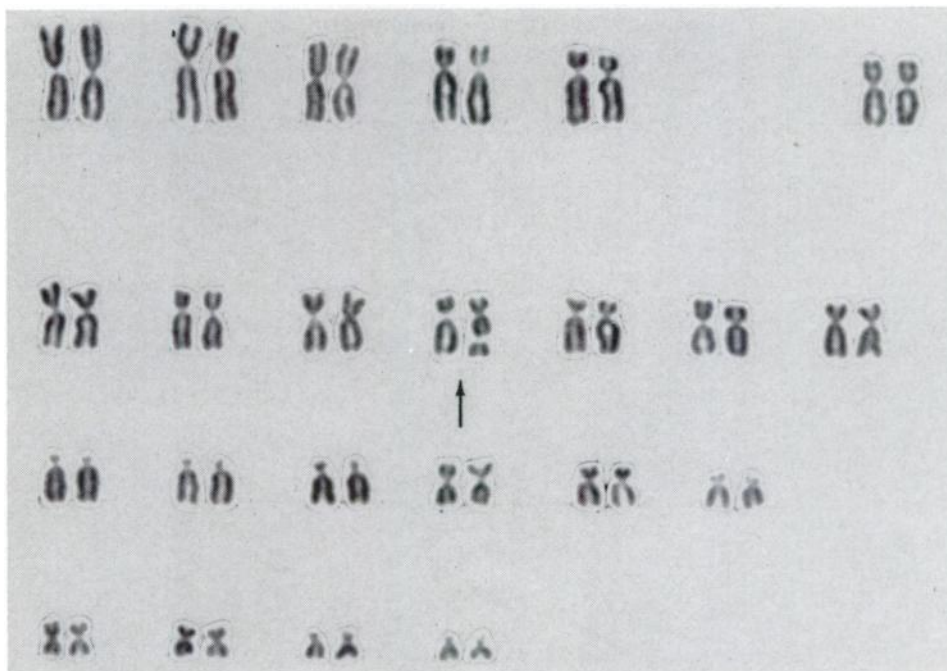


Fig. 1. Karyotype of the mother. Arrow points to a chromosome break.

105r/min, total dose per treatment 100r. Utilizing the radiation factors supplied by the respective physicians and converting these according to the tables in "Depth Dose Tables for Use in Radiotherapy," (1) we have arrived at the total skin dose of 1550r. If we take 3 cm as the average depth to the bone marrow and lymphoid tissue, the total dose will be reduced to 470 rads. All of the doses were estimated as being for soft tissue and; therefore, the dose to the bone marrow and lymphoid tissue had to be decreased by a factor of about 0.6. Multiplying 470 by 0.6 we arrived at a tentative total dose of 242 rads received by a portion of the bone marrow and lymphoid tissue.

She had a modal number of 46 chromosomes but examination of her karyotypes from three blood cultures revealed the presence of two types of chromosomal abnormality. The first type consisted of a break involving the same general area in the long arms of probably the same chromosome of Group C (Figs. 1, 2); the second type consisted of various aberrations (inclusive one dicentric) which involved randomly all chromosomes. The first type of lesion was several times more common, and we attempted to identify the affected chromosome in Group C. For this reason, measurements of arms and calculations of the centromere index in six karyotypes were carried out in accordance with the recommendation of the Denver Conference (2) and the values obtained were compared with the tabulated measurements of Penrose (3). The dimensions of the chromosomes with frequent break in the long arms were consistent with the presumed No. 9 chromosome; this finding was further strengthened by the presence of a marked secondary constriction in that chromosome (4). The secondary constrict-

tion, when present, was usually situated on the long arms close to the centromere, while the "weak" region, which was involved in breaks, was located at distal $\frac{1}{2}$ of the long arms.

Table I shows the results of chromosome examinations in the blood and skin cultures performed at different times. The rate of the abnormalities of the presumed chromosome No. 9 in all blood cultures was very high, ranging between 22.0 and 26.8 percent of the cells scored. The chromosome and chromatid aberrations seen in other chromosomes than the presumed No. 9 were distributed randomly; here the overall rate of chromosome-type aberrations alone was 3.5 percent, which is excessively high. According to Bender and Gooch (5) and Jacobs *et al* (6), the chromosome-type aberrations (dicentrics, rings, deletions

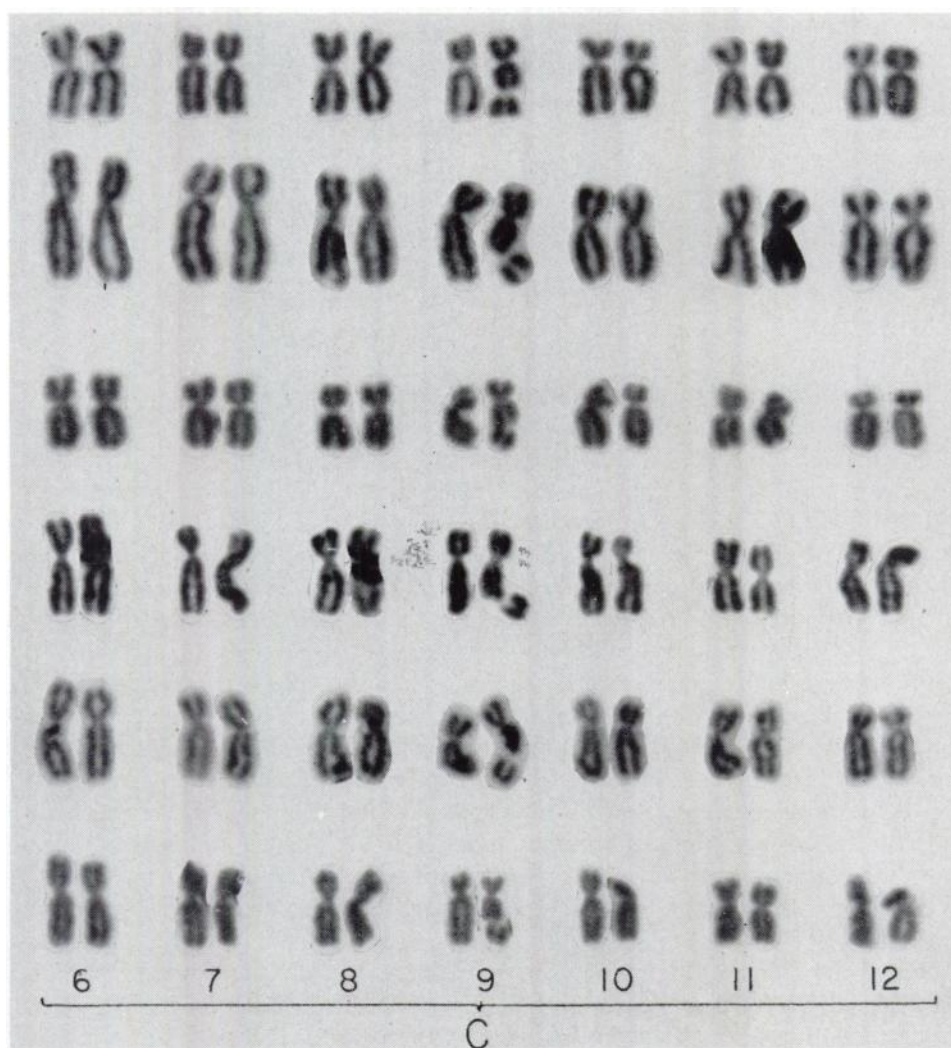


Fig. 2. A composite of chromosomes of Group C from six cells, Note deletion in the presumed No. 9 chromosome.

and translocations) do not exceed 1 percent in the normal population. The total rate of chromosome abnormalities involving the presumed No. 9 chromosome was five times higher than the rate of random aberrations present in the blood cultures of this female.

The percentage of mitoses with chromosomes of normal number and appearance was similar in all three blood cultures, and it was much lower than in normal controls. Since the incidence of cells with abnormal numbers of chromosomes in the present subject was about the same as in the general population (7), the relatively low frequency of euploid mitoses with normal chromosomes was solely the result of the high incidence of morphological abnormalities in her chromosomes.

In the skin culture the rate of chromosome-type aberrations was 6.0 percent and of the chromatid-type 8.8 percent. These rates are somewhat high, although they are within the range seen in transplants of fibroblastic cells of normal subjects (8, 9). There were no breaks involving two arms of the presumed No. 9 chromosome (Table I). In two mitoses there was one chromatid break and one chromatid gap each, involving the long arms of the presumed chromosome No. 9; however, the break was situated close to the centromere, unlike the lesions seen in the blood cultures, and the gap was close to the distal end.

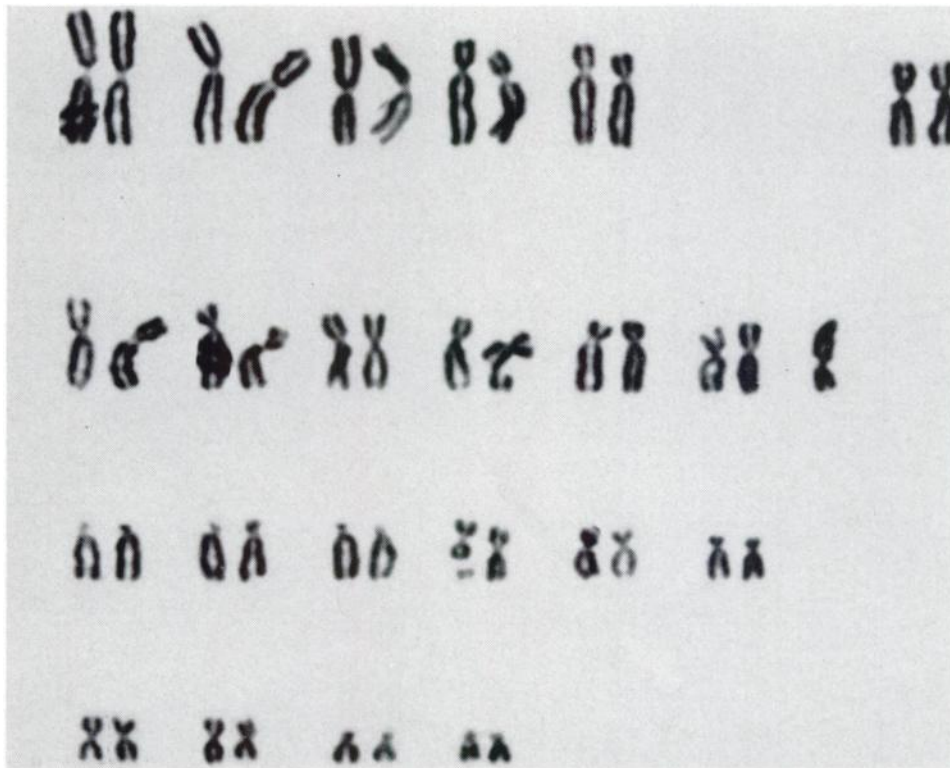


Fig. 3. Karyotype of a cell with 45 chromosomes. Note break involving the presumed chromosome No. 9 and chromosome No. 16.

TABLE I

Table I. Results of scoring chromosomes in three blood cultures and in a skin culture. The numbers in parenthesis in the first column refer to hypo- and hyper- modal chromosome numbers respectively. The dicentric chromosome (asterisk) was counted as two breaks.

Type of Culture and Date	No. Cells Scored	No. Euploid Cells with Normal Chromosomes	Abnormalities in Presumed Chromosome No. 9			Aberrations Involving Other Chromosomes than Pres. No. 9		
			Chromos. Break or Deletion	Chromatid Break or Gap	Total	Chromos. Break or Deletion	Chromatid Break or Gap	Total
1st Blood Culture 3/3/64	56 (2/1)	35 (62.5%)	12	3	15 (26.8%)	1	3	4
2nd Blood Culture 9/11/64	50 (1/0)	33 (66.0%)	10	1	11 (22.0%)	2	1	3
3rd Blood Culture 12/1/64	63 (2/1)	41 (65.1%)	13	3	16 (25.3%)	1 + 1* dicentric	2	5
Total	169 (5/2)	109 (64.4%)	35 (20.7%)	7 (4.1%)	42 (24.8%)	6 (3.6%)	6 (3.6%)	12 (7.1%)
Skin Culture 11/3/64	34 (3/0)	27 (79.4%)	0	2	2 (6.0%)	2	1	3 (8.8%)

COMMENT

The presence of a lesion in the same general area of long arms in the presumed chromosome No. 9 in about 24 percent of the mitoses in blood cultures is of considerable theoretical interest. About three fourths of the breaks affected both arms and one fourth of one arm. Since the rate of this abnormality persisted unchanged during repeated cultures, it is reasonable to propose that the patient has a clone of cells with a "weak" region in the presumed chromosome No. 9. The presence of this "weak" region is in some way responsible for the break of both arms in a proportion of mitoses and for one chromatid break or gap in the same general region in other cells of that clone.

It is conceivable that the abnormality originated sometime during the past 16 years when the patient has received x-ray treatments. Subsequently, the clone could have become well established in one of the blood forming tissues since the skin cultures are free of this particular abnormality. Clones of cells with abnormal chromosomes have been found in the bone marrow of irradiated mice (10). Nonetheless, another possibility of the origin of this lesion in the presumed chromosome No. 9 should also be considered. It is possible, but less likely, that she had this abnormality prior to the age of 16 years or even during fetal life. However, the chromosomes of her father were normal and her mother is deceased. Thus, no further light is shed on this assumption.

The presence of 3.5 percent of random chromosome-type aberration in short term blood cultures is in excess of what is seen in normal controls, and it is consistent with the findings detected in subjects who were exposed to ionizing radiation (11, 5). Also, the amount of radiation received by this woman to the bone marrow and lymphoid tissue approaches the doses which have been shown to be associated with chromosomal aberration (12, 13). Norman *et al* (14) found evidence of chromosomal aberration in radiation workers who received a lifetime cumulative dose of 10r. It remains unknown how much radiation, if at all, she might have received to her gonads and whether she has an increased rate of abnormal ova as compared to normal controls.

SUMMARY

Chromosome studies performed on the mother who gave birth to a dys-trophic and malformed child showed two types of abnormality. The first abnormality consisted of a clone of cells characterized by frequent breaks in the same general region of long arms in the presumed chromosome No. 9. Since this was observed in the repeated blood cultures but not in the skin culture, the clone is probably confined to the blood forming tissue. The second abnormality consisted of a moderately increased rate of chromosome-type aberrations occurring at random in all chromosomes. This woman received 1550 rads to the skin for treatment of eczematous dermatitis during the preceding 16 years, resulting in an estimated dose of 242 rads to portions of the bone marrow and lymph nodes. The type of random chromosome abnormality observed in her blood cultures has been reported previously to be associated with exposure to radiation. The aberrant cell clone may also have been induced by the same etiological factor.

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