

## Chromosome Studies on Marshall Islanders Exposed to Fallout Radiation

**Abstract.** *Cytogenetic studies of blood lymphocytes of Marshall Islanders, 10 years after their exposure to radiation from fallout in 1954, show chromosome-type aberrations in 23 of 43 exposed persons. Half the aberrations are of the exchange type. An unexpectedly large number of acentric fragments, but no exchange-type aberrations, appear in a few unexposed people on the same island.*

Chromosome aberrations in blood lymphocytes have been demonstrated in several population groups exposed to ionizing radiation, including patients during and after radiotherapy for ankylosing spondylitis (1) or malignant tumors (2), persons exposed during diagnostic procedures (3), and others exposed in the course of their work (4). Similar findings have been reported from individuals involved in radiation accidents (5) and in survivors of the atomic bombings of Hiroshima and Nagasaki in 1945 (6-8). One of the more interesting and possibly more significant points in all these studies was the observation that chromosome aberrations can persist in circulating lymphocytes for many years after the exposure. It seemed of interest to determine whether residual damage of this type also occurs in Marshall Islanders who were exposed to fallout radiation (9).

These people were accidentally exposed to radioactive fallout on Rongelap Island in 1954: the wind shifted unpredictably after the detonation of a high-yield nuclear device at Bikini, 160 km away; 64 inhabitants were exposed to 175 rads (estimated, whole-body) of gamma rays. Eighteen other Rongelap Islanders, away fishing on a neighboring island, were exposed to 70 rads (estimated). Exposure resulted in tem-

porary depression of blood cells, skin burns from beta radiation, and internal absorption of radionuclides, the most important being iodine and strontium. It was estimated that the thyroid glands of the adults received 150 rads—those of the children, as much as 1,000 rads—from absorbed radioiodine in addition to the whole-body exposure to gamma rays. Recent development of pathological changes in the thyroid is believed to have resulted from this exposure (10).

Chromosome preparations were obtained from 51 individuals during the annual medical review of the Marshall Islanders in 1964; 30 of them had been exposed to 175 rads; 13, to 70 rads of gamma rays. Eight Marshall Islanders who had escaped exposure were included for comparison. The exposed group comprised 20 males and 23 females, their ages ranging from embryo *in utero* to 67 years at the time of exposure. Twenty-one were under 20 and 22 over, with males and females about equal in number. The comparison group included three males and five females ranging in age from 10 to 71 years. Unfortunately it was impossible to examine more unexposed individuals matched to the exposed with respect to age and sex.

Cultures of peripheral blood lymphocytes were made by use of a modification of the method of Moorhead *et al.* (11); they were harvested at 48 and 72 hours (12). The slides were stained with aceto-orcein and examined by phase microscopy. Because comparison of the 48- and 72-hour cultures from five individuals showed no differences in aberration rates, all examinations but one were made on 50 cells of each individual from the 72-hour cultures. Altogether 2150 cells were examined from the 43 exposed subjects; 400 cells from the eight unexposed.

Aberrations were scored as follows: (i) aneuploidy, including polyploid cells, (ii) chromosome aberrations, and (iii)

chromatid aberrations. Karyotypes were made in cases where the counts were equivocal or where chromosomes of questionable morphology were seen. Only aberrations that were agreed on by several observers were included in the final tabulation. Additional karyotypes were made from eight euploid cells, showing no gross abnormalities, from each of four subjects showing more aberrations than most others; this was done in an effort to detect minor structural abnormalities such as small deletions or inversions that could easily escape notice under the microscope. Finally, bone-marrow preparations were made from two exposed subjects and one control; only one preparation from an exposed subject contained enough suitable cells in metaphase to warrant evaluation.

Proportionally the largest number of aberration-positive persons appeared in the low-exposure group (Table 1); we cannot explain this paradox and so treat all the exposed subjects as one group in comparing them with the unexposed (Table 1).

Chromosome-type aberrations were found in 23 of the 43 exposed people and in five of the eight unexposed. The high incidence of acentric fragments in the latter group was unusual and unexpected, and we cannot account for it. The total numbers of all types of chromosome aberrations in the two groups were 43 and 9—rates of 2.0 and 2.25 percent, respectively. Thus superficially there appears to be no difference between the two groups, but we should point out that in the exposed group half of all chromosome aberrations consisted of two-break aberrations such as dicentric chromosomes, translocations, and a ring, the remainder being acentric fragments. In contrast, no two-break aberrations were found in the unexposed group in which all aberrations were acentric fragments. Only seven of the exposed people resembled the controls in showing single-break aberra-

Table 1. Summary of cytogenetic findings in Marshall Islanders exposed to fallout radiation. Percentages appear in parentheses.

Exposure group (rads)	Subjects (No.)	Cells scored (No.)	Cells with $2n \neq 46$ (%)	Chromosome aberrations					Total chromosome breaks	Chromatid aberrations (No.)	
				Subjects affected	Fragments	Dicentrics	Rings	Translocations			Total
175	30	1500	10	12	11(0.73)	6(0.4)		5(0.33)	22(1.46)	33(2.2)	58(3.8)
70	13	650	8	11	10(1.53)	2(0.3)	1(0.15)	8(1.23)	21(3.23)	32(4.9)	35(5.3)
	43	2150	9	23	Totals for exposed subjects						
					21(0.93)	8(0.37)	1(0.04)	13(0.61)	43(2.0)	65(3.02)	93(4.3)
	8	400	5.5	5	Totals for unexposed subjects						
					9(2.25)				9(2.25)	9(2.25)	11(2.7)

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tions only; all others showed two-break aberrations either alone or in combination with fragments. The difference between the exposed and unexposed groups in the relative incidence of two-break aberrations and acentric fragments was significant at the 1-percent level ( $P < .01$ ).

Aberrations ranged from one to four per person. Sixteen of the 23 exposed and two of the five unexposed persons had multiple aberrations. In most instances no more than one aberration was noted per cell. Aneuploidy was higher in the exposed than in the unexposed group, but the difference was not significant; in most cells, aneuploidy was due to loss of chromosomes. The number of chromatid and isochromatid gaps and breaks, although somewhat higher in the exposed group, was within normal limits. Polyploid cells occurred with equal frequency in the two groups. No aberrations were found in the karyotypes that had been made from the 32 grossly intact cells from the four subjects showing relatively high rates of aberration. Likewise, no abnormalities were seen in 50 cells from the bone marrow of one of the exposed subjects.

We pointed out earlier that the proportion of aberration-positive persons in the low-exposure group was twice that among the highly exposed. We cannot explain this paradoxical effect, but we should mention that Bloom *et al.* also failed to detect a relation between chromosomally abnormal survivors in Hiroshima and Nagasaki and their estimated doses of radiation (8). Chromosome aberrations did not correlate with age or sex, nor could any relation be established between the occurrence of thyroid lesions and chromosome abnormalities.

We cannot account for the unusually high incidence of acentric fragments in the unexposed and their relative deficit in the exposed people. Radiation from ingested radionuclides is not likely to be a factor, since all Marshall Islanders have been exposed to low levels of residual radionuclides such as  $Cs^{137}$ ,  $Zn^{65}$ , and  $Sr^{90}$  since their return to Rongelap in 1957, and since body burdens of these elements were about the same in both groups. Although viruses

and some chemicals, in addition to ionizing radiations, are known to produce breaks in human chromosomes *in vivo* and *in vitro* (13), the kind and distribution of the fragments seen in the cells of the unexposed group do not suggest this origin. If these agents were implicated, it is not clear why they failed to produce a similar effect in the exposed people who have lived on the same island under identical environmental conditions since 1957.

Similar chromosome aberrations have been reported in the Japanese fishermen exposed to radiation from the same fallout (7); the incidence of aberrations, excluding aneuploid cells, was 2.1 percent and thus identical with our finding; most were two-break aberrations. The incidence of acentric fragments in a control population of Japanese studied by the same authors was 0.11 percent—20 times less than our finding among the eight unexposed Marshall Islanders. In a controlled cytogenetic study of sampled survivors of the atomic bombings of Hiroshima and Nagasaki, Bloom *et al.* have found exchange-type chromosome aberrations in 33 persons—35 percent of 94 survivors examined 20 years after exposure (8); the incidence of aberrations was 0.6 percent in the exposed group—less than half our finding among the Marshall Islanders. Bloom *et al.* found only a single acentric fragment in 8847 cells from the 94 controls, an incidence of 0.01 percent; for the 33 aberration-positive individuals, the dose ranged from 237 to 891 rads, and for none of the entire group was it less than 200 rads. All persons examined were 30 years of age or younger at the time of the bombings.

The results of our study demonstrate that a small but significant number of chromosome aberrations persists in blood lymphocytes of some Marshall Islanders 10 years after exposure to fallout radiation. The conclusion that at least some of these aberrations were caused by the radiation, and not by other factors, rests on the finding that exchange-type aberrations were found only in the exposed people and not among the controls.

The biologic significance of persistent chromosome aberrations in blood lym-

phocytes of hematologically normal persons many years after exposure to ionizing radiation is not known. In particular, any role that aberrations of this type may play in the pathogenesis of radiation-induced leukemia can only be surmised at present. It is problematical and indeed doubtful whether chromosome aberrations in lymphocytes can serve to indicate abnormalities in other tissues, except perhaps inferentially. Indeed, of the ten individuals who have developed thyroid lesions since our examinations were made, only three show double-break aberrations.

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