Experimental Section

Gerontology

Gerontology 1998;44:272–276

Received: August 25, 1997
Accepted: November 15, 1997

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Effect of a Continuous Gamma Irradiation at a Very Low Dose on the Life Span of Mice

Abstract

Background: There is epidemiological evidence that suggests there are beneficial effects of ionizing radiation at low doses. Some experimental studies confirmed this hormetic effect with doses of about 1 cGy/day, but no data concerning very low dose rates are available. **Objective:** The aim of this study was to determine the life span of mice exposed to very low doses of ionizing radiation. **Methods:** Six hundred female C57BL/6 mice, 1 month old, were exposed to chronic gamma irradiation at very low dose rates of 7 or 14 cGy/year. These doses are about 25 or 50 times higher than background, but much lower than the doses of about 1 cGy/day used in previous experiments. Three hundred mice living in the same room were used as controls. **Results:** The life span, after the beginning of the experiment, determined by the survival time of 50% of each population, is increased in irradiated mice: 549 days in controls, 673 days in both irradiated groups. The differences are significant between the control and the irradiation mice. Differences between mice irradiated with 7 or 14 cGy are not significant. **Conclusions:** These results confirm the possibility of a nonharmful effect (hormesis) of ionizing radiation. They demonstrate that the paradigm, which states that low-dose effects can be predicted high-dose effects, cannot be systematically applied in radiation biology in general and gerontology in particular.

Key Words
Ionizing radiation
Low doses, ionizing radiation
Life span

Introduction

It is well known that exposure to ionizing radiation can induce a shortening of the life span in man and animals. However, the survival time can be prolonged after exposure to low doses. This effect was reported, for the first time, by Lorenz et al. [1] in the 1950s: mice exposed to 0.11 rad for 8 h/day had a longer mean survival rate, nearly 2 months. According to Sacher and Grahn [2], exposure to 0.4 or 5 cGy/day of ^{60}Co gamma rays results in about a 10% increase of the life span. Similar results were reported in mice exposed to 1 cGy/day for many generations [3] or after exposure to 0.8 cGy/day for 1 year [4]. An increased life span was also noted in 2-month-old mice continuously irradiated at 0.7 or 6 cGy/day [5].

More recently, it was shown that the mean survival time was significantly prolonged from 283 ± 3 days in controls to 316 ± 10 days in mice irradiated with 15-cGy X-rays twice a week [6]. With X-ray or neutron single irradiation in mice performed on days 7 or 21 of age, survival

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0304–324X/98/0445–0272$15.00/0

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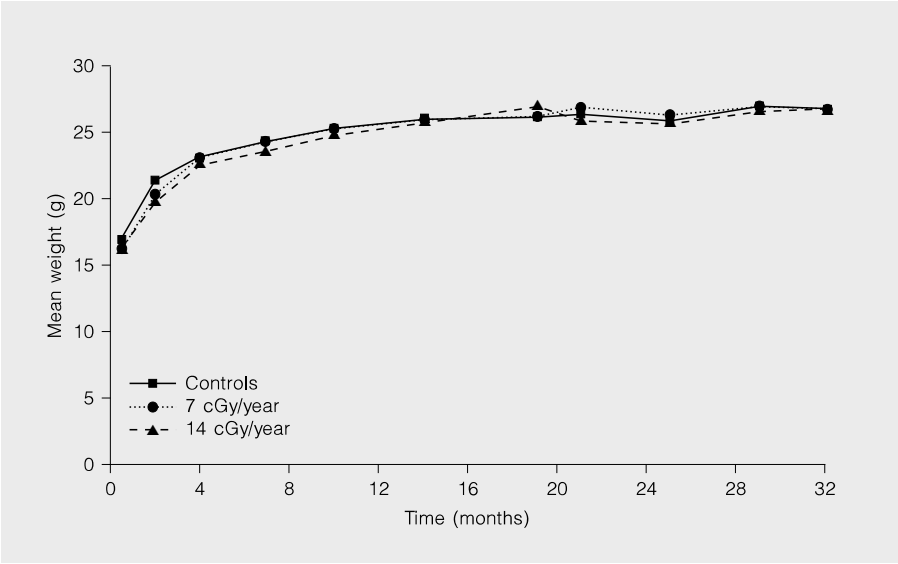

Fig. 1. Mouse weights from the beginning of the experiment to animal death in the different groups. No statistical difference was observed between groups.

was reduced with high doses but increased in groups receiving the lowest doses (0.125 Gy neutrons or 0.5 Gy X-ray) [7]. It can be pointed out that an increase in life span occurred, in most experiments, when a chronic irradiation was performed at a dose rate of about 1 or several cGy/day.

Over the past few years, we have conducted in our laboratory large-scale experiments in single-cell organisms of the effects of background and very low dose radiation. In particular, we have demonstrated that ionizing radiation, under these conditions, can stimulate cell proliferation [8] and affect the clonal life span in *Paramecium tetraurelia* [9].

Based on this research, the main objective of the present study was to determine if exposure to very low doses, much lower than those used in previous work, can result in changes in life span. This paper presents the effects on the life span in mice of a whole-body gamma chronic irradiation exposure at a dose rate of 7 or 14 cGy/year.

Materials and Methods

C57BL/6 female mice, 3 or 4 week old, were purchased from Iffa Credo (l'Arbresle, France). They were kept in a room lighted from 07.00 to 19.00 h and maintained at a temperature of 20–22°C. The total air volume was renewed at a rate of 1,500–2100 m^3/h. A standard diet was provided ad libitum. The animals were housed 30 per cage and kept until spontaneous death.

Irradiation was given by thorium nitrate in air-tight plastic bags, placed beneath the floor of the cages. Dosimetry was performed using Fli detectors kept for 4 weeks at the cage floor level. Measurements were done at the Commissariat à l'Energie Atomique (Fontenay-aux-Roses. France).

Nine hundred animals were divided into three groups: (1) 300 as controls; (2) 300 irradiated at a dose rate of 7 cGy/year, and (3) 300 irradiated at a dose rate of 14 cGy/year. The mice were weighed and checked for death every day except Sunday.

Statistical analysis was performed as follows [10–12]: (1) Comparison by χ^2 test of the survival frequencies observed on twelve different dates. (2) Comparison of the regressions between survival rate and time by means of covariance analysis after probit transformation. This provides a good linearization of the relationship before and after nonparallelism test of the regression straight lines. (3) Estimation of the survival time of 50% of the population by means of two methods of weighted regression after probit and logit transformation of the survival rate (computation which can be compared to the lethal dose 50; LD_{50}).

Results

Figure 1 shows the weights of mice from the beginning of the experiment to the death of most animals. The weight was not measured during the last weeks, due to the too low number of mice still alive. The growth rates remained unchanged in irradiated mice.

As shown in figure 2, it is obvious that the survival capacity is higher in both irradiated groups than in controls. The survival curves can be fitted by a logistic model such as the Nelder curve. Comparison by χ^2 test of the survival frequencies observed on 12 different dates demonstrates that differences between irradiated mice and controls are significant (table 1).

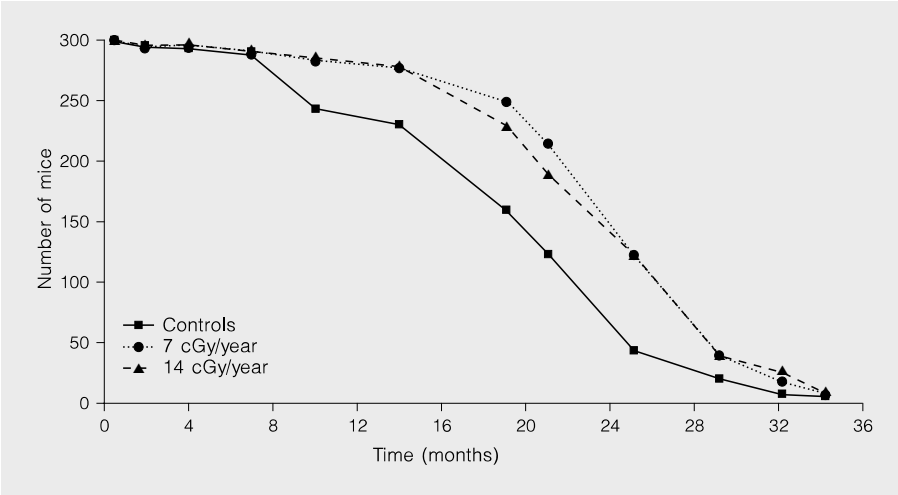

Fig. 2. Survival curves of the different groups. The life span is higher in both irradiated groups than in the control group ($p < 0.01$). Differences between the two irradiated groups are not significant (x = time in months from the start of the experiment; y = number of surviving mice).

Table 1. Comparison by χ^2 test of the survival frequencies on 12 different dates (ranging from the 1st to the 34th month) in the different groups of mice

Groups	χ^2	Degree of freedom	Probability	
			%	p
7 vs. 14 cGy/year	5.92	11	87.93	NS
7 cGy/year vs. controls	71.40	11	≤0.01	≤ 0.001
14 cGy/year vs. controls	65.12	11	≤0.01	≤ 0.001
7 vs. 14 cGy/year vs. controls	88.61	22	≤0.01	≤ 0.001

NS = Not significant.

On the other hand, relationships between survival rates and time can be linearized by means of probit or logit transformation of survival rates. The comparison of the regression survival rate/time by covariance analysis is shown in table 2.

Life lengthening in irradiated mice is shown in table 3 which indicates the survival time (days) of 50% of the population (ST_{50}). ST_{50} values are very similar using weighted probits or weighted logits (table 3).

Discussion

Exposure to chronic low-dose gamma irradiation results in a significantly increased life span expressed by the survival time of 50% of the population. The response is the same in mice exposed to the two dose rates.

Changes in longevity can be ascribed to radiation, as besides the irradiation, the three groups lived in an identical environment.

The mechanisms for the increased life span in mice exposed to very low doses are unknown. We could assume that radiation can have a protective effect against the incidence of neoplastic or nonneoplastic diseases. This mechanism can be involved in strains in which death of most animals is due to carcinomas or leukemia. In the AKR strain, in which about 80% of males die of lymphoma, the prolonged life span observed in mice irradiated with 5 or 15 cGy, respectively, three times or twice a week, appears to be related to a reduced incidence of lymphomas [6]. In C57BL/6 mice, the incidence of tumors is very low, and, in our experiments, tumors were not detected in both control and irradiated mice. Furthermore, the influence of irradiation could be due to a delay in the aging process or to a lower incidence of infectious diseases. Indeed, in C57BL/6 male mice, small doses of X-rays (0.5 Gy) or 3.1 MeV neutrons (0.125 Gy), which result in an increased life span, might reduce and/or delay nonneoplastic lung diseases [7].

Table 2. Comparison of the regression between survival rate and time by covariance analysis after probit transformation

Groups	Test of parallelism		Comparison of mean values		
	probability %	F (variance ratio)	degrees of freedom	probability %	p
7 vs. 14 cGy/year	77.23 (NS)	0.49	1–21	49.95	NS
7 cGy/year vs. controls	14.00 (NS)	6.93	1–21	1.49	≤ 0.05
14 cGy/year vs. controls	10.02 (NS)	21.67	1–21	≤0.01	≤ 0.001
7 vs. 14 cGy/year vs. controls	25.32 (NS)	7.15	2–32	≤0.27	≤ 0.01

NS = Not significant.

In both cases, the low-dose effects could be due to a radiation-induced stimulation of immune functions. Indeed, with high doses, the immune system can be activated: 20 Gy of gamma ^{60}Co rays induces an enhanced interleukin 2 production by human T lymphocytes [13]. In mice exposed to 75 mGy of X-rays, the T cell function was increased by 212% when compared to controls [14]. In China, where human populations are exposed to a high level of background radiation, the reactivity of lymphocytes, measured by [^{3}H]thymidine incorporation, is higher than in populations living in areas with a background radiation three times lower [15]. Furthermore, in the previous work [16], we have reported an enhancement in immune reaction. This is expressed by changes in cell populations of the thymus and the spleen of mice exposed to a chronic gamma irradiation at a dose rate of 74 mGy/year [16]. These results are in good agreement with those of previous investigations [17] which have shown a higher proliferation of splenic T cells in mice exposed to 20 days of gamma irradiation at doses from 5 to 40 mGy/day. Additional experiments are in progress in our laboratory in order to confirm this assumption.

On the other hand, changes in the life span after exposure to very low doses could be considered adaptive responses [18]: small doses can stimulate the DNA repair process and increase the resistance to high doses given a few hours later. In our experiment, changes in life span could be related to an enhancement in the repair of damaged DNA related to aging. Supporting this hypothesis is evidence of stimulated expression of a number of genes even with doses as low as 0.06 Gy [19].

Furthermore, the increase in life span reported in this paper supports the hormesis concept, i.e., there is a possible nonharmful effect of ionizing radiation [20]. Radiation hormesis is currently reported after exposure to low doses, about 1 Gy. However, it can be observed with much lower doses and, in particular, with background radiation: in paramecia and a blue alga, *Synechoccocus lividus*, protection against natural ionizing radiation is followed by a lower cell growth rate [8]. The results presented here show that hormesis, induced by very low doses, can also be observed in more complex organisms. Furthermore, they demonstrate that the radiation paradigm [21], which states that low-dose effects can be predicted from those observed after high-dose exposure, cannot be systematically applied in radiobiology and gerontology.

Table 3. ST_{50} values

	ST_{50}	95% confidence intervals of ST_{50}
Weighted probits		
Controls	549.6	536.9–562.3
7 cGy/year	673.1**	659.8–686.4
14 cGy/year	673.6**	660.7–686.5
Weighted logits		
Controls	553.8	540.8–566.7
7 cGy/year	684.6**	671.7–697.6
14 cGy/year	680.7**	668.1–693.4

ST_{50} values are similar using weighted probits or weighted logits: ** $p \leq 0.01$.

Acknowledgements

We thank G. Gasset and S. Cazeaux for their scientific and technical assistance. This study was supported by a research grant from Electricité de France (EDF).

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