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Life Shortening in BALB/c Mice following Brief, Protracted, or Fractionated Exposures to Neutrons[1,2]

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The effects of acute, protracted, or fractionated exposures to fission neutrons on survival times of female BALB/c mice were examined and compared. Mice were given single, brief exposures or exposures given in equal fractions at either 1- or 30-day intervals to doses of 0, 2.5, 5, 10, 20, 50, and 200 rad at the Health Physics Research Reactor (HPRR) or protracted exposures at rates ranging from 0.1 to 10 rad/day using a moderated ^{252}Cf source to doses of 0, 2.5, 5, 10, 20, and 40 rad. The ^{252}Cf source was moderated to have a similar spectron to that of the HPRR facility. After single or fractionated exposures the extent of life shortening increased rapidly over the 0–50 rad range and then began the plateau. No simple model adequately described the dose response over this entire dose range. Over the 0–50 rad dose range for exposures at the HPRR and over the 0–40 rad dose range for protracted exposures the dose response could be adequately described by either a linear model or a square root of the dose regression model except when the dose was fractionated using a 30-day interval. In this instance a linear model provided an adequate fit while a square root of the dose model could be rejected. No increase in effectiveness after fractionation or protraction was observed for neutron-induced life shortening at doses below 50 rad, while at 50 and 200 rad an increase in effectiveness was observed in this and in previous studies. These data were interpreted to suggest that in the dose range below 20–40 rad the dose–effect curve for life shortening may be linear and begins to flatten at higher doses rather than continuously bending at low doses.

INTRODUCTION

Among the major unresolved issues in neutron radiobiology are the following. In animals given total-body exposures, what is the precise shape of the dose–response curve for late effects such as life shortening or tumor incidence in the low-dose range? Are fractionated or protracted exposures more effective in producing late effects than single brief exposures, and is there evidence for an effect of dose rate with protracted exposures?

There seems to be general agreement that for some tumors in mice (*1–4*) and for life shortening in mice (*5, 6*) [see also a reassessment of earlier studies (*6*)] the dose–effect curve rises rapidly in the low-dose range and then begins to flatten at a dose

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of around 50 rad. In many cases, where an extended dose range is considered, the curve is reasonably well approximated as a function of the square root of the neutron dose. It is not clear, however, whether the curve continuously bends in the low-dose range. An alternative hypothesis is that the curve is linear up to a level of between perhaps 20 and 50 rad and then begins to plateau as some "saturation" level is reached. Choosing between these two hypotheses is of considerable theoretical as well as practical importance. If, as is now commonly believed, the initial slope of dose–effect curves for many responses to low-LET radiation is linear (*7*), and if the true shape of the neutron curve is a function of the square root of the dose, then it follows that the biological effectiveness of neutrons relative to low-LET radiation will continue to increase systematically as the neutron dose is decreased and will attain very high values in the range of interest for considerations of human radiation protection. On the other hand, a linear neutron slope combined with a linear slope for low-LET radiation will lead to a limiting and constant value for relative biological effectiveness (RBE).

In practice it has usually proved difficult to choose between these alternative shapes on the basis of goodness of fit to the data. In an earlier study on life shortening (*5*) we showed that a linear slope provided an acceptable fit in the range of 0–47 rad in RFM female mice and in the range of 0–20 rad in BALB/c female mice. In both cases, the square root function could be rejected ($P < 0.05$). We previously reported (*1*) that, in the dose range of 0–47 rad, the corrected incidence of thymic lymphoma in RFM mice increased linearly with dose ($P > 0.01$). We did not attempt a fit of the square root function at that time. Subsequently we have found that the square root model can be rejected ($P < 0.05$). More recently Ullrich (*4*) has been unable to reject either of the alternative models for fitting mammary or lung carcinoma incidence in BALB/c mice in the dose range below 50 rad. Similarly, Thomson *et al.* (*8*) were unable to choose between the alternatives (when weighted regression lines were used) for life shortening in male or female $B6CF_1$ mice in the dose range of 20 rad or less.

The question of whether fractionated or protracted exposure is more effective than a single brief exposure has also not been entirely resolved despite its importance in contributing to an understanding of neutron radiobiology and in setting radiation standards. There is consistent evidence that at relatively high total doses, fractionation or protraction is more effective. We previously showed (*5*) that a continuous exposure at 1 rad/day to a total of 188 rad produced significantly greater life shortening in RFM and BALB/c female mice than did the same dose delivered in one session at 25 rad/min. There was also a significantly higher incidence of thymic lymphoma in the RFM mice with protracted exposure to this dose level (*1*). In the studies reported by Thomson *et al.* (*6*) the fractionated exposures produced significantly more life shortening at doses of 60 or more rad in $B6CF_1$ male mice and at doses of 160 or more rad in the female mice. At lower doses there were no significant differences. We have found (*9*) that 50 rad given in two fractions 30 days apart caused a higher incidence of lung and mammary carcinomas in BALB/c female mice than did a single exposure. Again, at lower doses there were no significant differences. It appears, then, that fractionated or protracted exposures are more effective than single brief exposures when relatively high total doses are used. It is not at all clear, however, that this effect holds in the dose range below 50 rad.

We are unaware of data from irradiated animals exposed continuously at various dose rates to determine whether neutrons will show a dose–rate effect at low total doses. In the present paper we report data on survival times in over 6600 BALB/c female mice given single brief exposure to neutrons, exposures given in equal fractions at either 1- or 30-day intervals, and continuous exposures at rates ranging from 0.1 to 10 rad/day, along with data from sham-irradiated (control) mice. The detailed pathology obtained on these animals will be reported later.

MATERIALS AND METHODS

Radiation. The single brief exposures and the fractionated exposures were made using the Oak Ridge Health Physics Research Reactor (HPRR). A description of the reactor and methods of dosimetry were provided previously (*5*). In essence the neutron energies were those of a slightly degraded ^{235}U-fission spectrum with a neutron-to-gamma-ray dose ratio at the point of exposure of the mice of 7:1. As in previous studies (*5*) and for reasons discussed below we ignored the approximately 15% dose contribution from the gamma rays. Dose rates ranged from 1 to 25 rad/min depending on the total dose desired. This variation was due to the fact that we avoided exposure times of less than 1 min.

The continuous (20 hr/day) exposures were made using a 5.4-mg ^{252}Cf source housed in a lightly constructed (frame) facility. As with the source used previously (*5*), we surrounded this source with a sphere of depleted ^{238}U with a wall thickness of 6.35 cm. This sphere was used to degrade the neutron energy spectrum to make it comparable to the HPRR spectrum. Dosimetry was performed essentially as previously described (*5*). The principal differences in the radiation field from that used previously were due to the elimination of the massive concrete shielding. This resulted in a decrease in the low-energy component of the neutron spectrum and a marked reduction in the gamma-ray contamination. Except at energies above 2.5 MeV where the ^{252}Cf field showed a relative deficit, the spectra for the HPRR and the ^{252}Cf source were similar. The neutron-to-gamma-ray dose ratio varied with distance from the source. At the distances at which most exposures were made (approximately 1 and 3 m) the gamma-ray component consisted of 6 to 13% of the dose. Only at a distance of 9 m (where one group was exposed) did the gamma-ray contamination exceed that for the HPRR, reaching a level of about 25%. Again we ignored any possible contribution from the gamma-ray dose, and the doses shown represent neutron doses. Exposures of the mice were made as described previously (*5*). As the source decayed (2.646 year half-life) the distances from source to cages were adjusted.

The gamma-ray contributions to dose were ignored for the following reasons. With the HPRR the gamma rays contributed about 15% of the total dose. At the highest dose of 200 rad of neutrons there would be an additional 30 rad from gamma rays. If we assume any reasonable value for the RBE of neutrons it becomes apparent that the contribution of gamma rays to the observed effect must be very small. For example, if we assume an RBE of 10, then the 30 rad of gamma rays would contribute an effect equal to 3 rad of neutrons. This lies within the errors associated with the dosimetry. Similarly, with the ^{252}Cf source the maximum gamma-ray contribution was 25% at a neutron dose of 10 rad delivered at 0.1 rad/day. Thus at this dose there was a 2.5 rad gamma-ray dose. If the RBE of neutrons is 10 then the effect anticipated

would be equal to that from 0.25 rad of neutrons. Accordingly, we felt justified in ignoring the gamma-ray component of dose.

Statistical methods. Mean ages at death (which we equate to mean survival times) and their standard errors were calculated for each experimental and control group. In determining whether, at equal dose levels, the mean survival times differed significantly we used the standard t test for comparisons between two groups and the F test when three or more groups were compared. Throughout we have rejected the null hypothesis at $P < 0.05$.

In fitting weighted regression lines to the data we used two alternative regression models. These were

$$\text{Linear model} \qquad y = a + bX,$$

$$\text{Square root model} \qquad y = a + b(X^{0.5}),$$

where y = mean survival time in days and X = dose in rad. In all cases we used the observed survival times rather than days of life shortening. This enabled us to use the control values as an experimental point. As we pointed out previously (*5*), the standard error (and variance) of life shortening in controls is zero and the control value cannot be properly weighted. Further, the values for life shortening, as opposed to mean survival times, are not independent since they are derived by subtracting from a constant (the control survival time). In fitting regression lines to mean survival times, the weight given each mean value should be proportional to the amount of information in or the precision of the observation. The precision of the estimate of each mean is directly proportional to the sample size (n_i) and inversely proportional to the sample variance (σ_i^2) (*10*). The weighting factor (w_i) for each mean is therefore $w_i = n_i/\sigma_i^2$. This is formally the equivalent of the inverse of the squared standard error of the mean. We weighted our regression lines in this manner. For a weighted regression line the weighted residual sum of squares or the sum of the weighted squared deviations from regression is distributed as χ^2 (*11*) and goodness of fit can be evaluated from standard χ^2 tables with $k - a$ degrees of freedom where k = no. of data points and a = no. of constants in the equation. We used this method to test for goodness of fit or, in other words, to evaluate the probability that the deviations from the regression were due to chance random sampling variation.

Mice. Barrier-maintained, specific-pathogen-free, female BALB/c/An N Bdf mice were used in these experiments. A description of animal husbandry practices and radiation exposure methods has been provided previously (*5*). The mice were 12 weeks old at the time radiation exposures were begun. In most cases the mice were killed when moribund. This procedure enabled us to obtain autopsy information on over 98% of the animals. Basically there were two experiments which were run consecutively. In the first experiment the mice were exposed to single or fractionated (two equal fractions) doses of neutrons delivered at a high dose rate from the Health Physics Research Reactor. A large group of controls was run concurrently. The second experiment consisted of continuous exposure at various dose rates to neutrons from the ^{252}Cf source. A small control group was used in this experiment inasmuch as we expected the control survival time would not differ from that found in the first experiment. As will be seen, this expectation, unfortunately, was not realized.

Sample sizes, radiation doses, fractionation schedules, mean survival times, and

days of life-shortening for the first experiment are shown in Table I. Table II shows sample sizes, radiation doses and dose rates, mean survival times, and days of life shortening for the second experiment.

RESULTS

It is apparent from Table I that at the highest dose level employed (200 rad), there was a plateauing of the effect on mean survival times and therefore on the extent of life shortening. This flattening of the curve has been reported previously (*5, 6*). Figure 1 illustrates the effect in mice receiving a single exposure to neutrons from the HPRR. Since the issue of principal interest is the shape of the curve in the low-dose range we concentrated our analysis on the lower end of the curve.

It is also apparent from a comparison of Tables I and II that survival times for control mice and for mice irradiated at equal total doses differed significantly in the two experiments. For reasons unknown to us the mice in the second experiment lived longer on the average and data on survival times from the two experiments could not be pooled. A separate analysis of the two experiments was therefore required.

We first examined the question of whether there were significant differences in mean survival times, at equal total doses, among the mice exposed at the HPRR to single or two fractionated exposures (Table I). We found no significant differences

TABLE I

Fractionation Schedule, Total Dosage, Sample Size, Mean Survival Time, and Life Shortening in Mice Exposed to Neutrons from the Health Physics Research Reactor

Interval between two equal fractions	*Total dose (rad)*	*No. mice*	*Mean survival time (days) ± SE*	*Life shortening (days) ± SE*
—	controls	556	736.3 ± 7.38	—
0 (single exposure)	2.5	185	726.8 ± 13.02	9.5 ± 14.97
	5	189	715.0 ± 11.98	21.3 ± 14.07
	10	185	696.1 ± 11.97	40.2 ± 14.06
	20	182	677.6 ± 10.03	58.7 ± 12.45
	50	189	646.9 ± 10.80	89.4 ± 13.08
	200	185	628.3 ± 10.48	108.0 ± 12.82
1 day	2.5	192	686.3 ± 13.24	50.0 ± 15.16
	5	190	727.4 ± 12.13	8.9 ± 14.20
	10	189	707.4 ± 11.84	28.9 ± 13.95
	20	186	693.8 ± 9.67	42.5 ± 12.16
	50	187	634.0 ± 11.00	102.3 ± 13.25
	200	185	612.9 ± 12.49	123.4 ± 14.51
30 days	2.5	189	738.8 ± 11.42	−2.5 ± 13.60
	5	188	739.1 ± 11.29	−2.8 ± 13.49
	10	187	719.3 ± 11.70	17.0 ± 13.83
	20	181	672.5 ± 10.68	63.8 ± 12.98
	50	191	618.2 ± 11.12	118.1 ± 13.35
	200	185	552.4 ± 11.64	183.9 ± 13.78

TABLE II

Radiation Dose, Dose Rate, Sample Size, Mean Survival Time, and Life Shortening in Mice Exposed to Neutrons from a ^{252}Cf Source

Total dose (rad)	*Dose rate (rad/day)*	*Duration of exposure (days)*	*No. mice*	*Mean survival time (days) ± SE*	*Life shortening (days) ± SE*
Controls	—	—	191	785.0 ± 11.10	—
2.5	1	2.5	200	770.3 ± 11.61	14.7 ± 16.06
	10	0.25	198	754.1 ± 11.16	30.9 ± 15.74
5	1	5.0	196	758.1 ± 10.81	26.9 ± 15.49
	10	0.5	198	738.6 ± 11.80	46.4 ± 16.20
10	0.1	100	197	734.6 ± 11.67	50.4 ± 16.11
	1	10	193	748.0 ± 10.53	37.0 ± 15.30
	10	1	197	733.7 ± 10.49	51.3 ± 15.27
20	1	20	395	702.2 ± 8.49	82.8 ± 13.97
	3	6.67	197	726.1 ± 9.99	58.9 ± 14.93
	10	2	192	719.5 ± 12.14	65.5 ± 16.45
40	1	40	198	671.8 ± 10.89	113.2 ± 15.55
	10	4	194	660.2 ± 11.48	124.8 ± 15.97

at doses of 5, 10, or 20 rad. At 2.5, 50, and 200 rad the survival times differed significantly ($P < 0.05$). Examination of the data at the 2.5-rad point indicated that the difference was due to an unusually short survival time in the group receiving a split dose at a 1-day interval. The survival time was shorter than those seen at the 5, 10, and 20-rad points. Examination of the pathology in this group indicated an unusually high incidence and/or earlier onset of tumors of the reticular tissues. No other group showed this effect. For this reason and because of the marked deviation from survival times in adjacent dose groups we felt justified in concluding that it

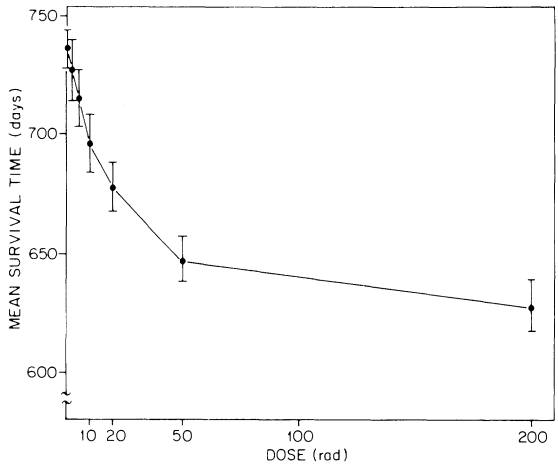

FIG. 1. Mean survival times (with standard errors) of mice as a function of dose of fission neutrons delivered in a single exposure at a high dose rate.

represented an unusual sampling variation and eliminated this one dose group. The remaining two groups at 2.5 rad did not differ significantly. Because the survival times did not differ significantly at 2.5 (after eliminating one group), 5, 10, and 20 rad we then pooled the data at each dose level to establish mean survival times with greater precision. The pooled data are shown in Table III. At 50 and 200 rad the survival times differed significantly and at both dose levels the ordering was the same, namely, the 30-day split caused the most life shortening followed by the 1-day split and the single dose in that order.

In the case of mice receiving protracted exposures (Table II) we found no significant differences in survival times at equal dose levels regardless of the dose rates utilized. The dose rates in most cases varied by a factor of 10 but in the case of the 10-rad group the variation amounted to a factor of 100. This finding supports the conclusion for the split-dose group that at total doses of less than 50 rad it made little if any difference how the doses were spaced in time. Again, to establish mean survival times with greater precision, we pooled the data at equal dose levels and recalculated survival times. The results are shown in Table III.

We next estimated the goodness of fit of the two alternative regression models to the mean survival times. The models considered were the linear model where survival times decrease as a linear function of dose and the square root model where survival times decrease as a function of the square root of the dose. In the case of single brief exposures or equal fractions delivered at intervals of 1 or 30 days we rejected both models when they were applied to the full dosage range of 0 to 200 rad ($P < 0.01$). When we restricted the dosage range to 0 to 50 rad, either model gave an acceptable fit to the data for single exposures or for fractions delivered at a 1-day interval. P values were >0.2 and >0.9, respectively, for the linear model and >0.9 and >0.1 for the square root model. When the fractions were delivered at a 30-day interval the linear model gave an acceptable fit ($P > 0.3$) but the square root model could be

TABLE III

Pooled Data (See Text) for Mice Exposed to Neutrons from the HPRR[a] or a ^{252}Cf Source

Radiation source	*Total dose (rad)*	*No. mice*	*Mean survival time (days) ± SE*	*Life shortening (days) ± SE*
HPRR[a]	Controls	556	736.3 ± 7.38	—
	2.5	374	732.9 ± 9.29	3.4 ± 11.86
	5	567	727.2 ± 6.83	9.1 ± 10.06
	10	561	707.6 ± 6.52	28.7 ± 9.85
	20	549	681.3 ± 5.86	54.9 ± 9.42
^{252}Cf	Controls	191	785.0 ± 11.10	—
	2.5	398	762.2 ± 8.32	22.8 ± 13.87
	5	394	748.3 ± 8.02	36.7 ± 13.69
	10	587	738.7 ± 6.31	46.3 ± 12.77
	20	784	712.4 ± 5.64	72.6 ± 12.45
	40	392	666.0 ± 7.91	119.0 ± 13.63

[a] Health Physics Research Reactor.

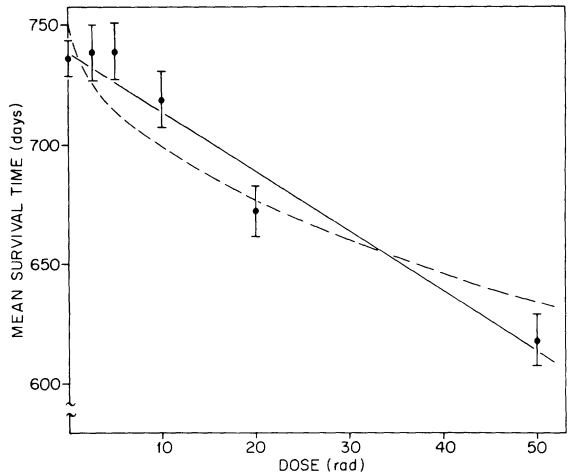

FIG. 2. Mean survival times (with standard errors) as a function of total dose for mice exposed to fission neutrons delivered in two equal fractions separated by 30 days. Solid line is the least squares fit for the linear model. Dashed line is for the square root of dose model.

rejected ($P < 0.01$). This represented the only case in which we had a basis for choice between models. Data on mean survival times and the two regression lines for the 30-day fractionation groups are shown in Fig. 2. This figure illustrates well the difficulty in choosing between models in a restricted dose range, the differences in the lines being small relative to the size of the standard errors. When we further restricted the dose range, e.g., 0–20 rad, either function gave an acceptable fit.

Inasmuch as we were able to pool the data for groups exposed to 20 rad or less at the HPRR (Table III), we tested the two models on the pooled data. Because of the larger sample sizes these means can be considered to have been established with greater precision. We were unable to reject either model although the linear model gave a somewhat better fit ($P > 0.9$) than the square root model ($P < 0.1$). Mean survival times as a function of dose for the pooled data are shown in Fig. 3. Both the linear regression line and the square root function are shown in this figure.

With protracted exposures to the ^{252}Cf source we were unable to reject either model over the dose range studied. As indicated earlier we found no significant differences in survival times at equal dose levels regardless of the differences in dose rates. Data on mean survival times as a function of dose for these pooled data (Table III) are shown in Fig. 4. Again we have shown both the linear regression relationship and the square root of the dose function.

The slopes for the linear functions shown in Figs. 3 and 4 are -2.87 ± 0.157 for the fractionated exposure (Fig. 3) and -2.63 ± 0.231 for the protracted exposure (Fig. 4). These slope constants are obviously not significantly different, again indicating that in the low-dose range it does not seem to matter how the dose is distributed in time. If the dose–effect relationship is truly linear in this dose range then it follows that life shortening amounts to about 2.7 days/rad at low doses. We previously (5) estimated life shortening from gamma rays in this strain at about 0.26 to 0.39 days per rad depending on dose rate. This would suggest an RBE of around 10 in the low-

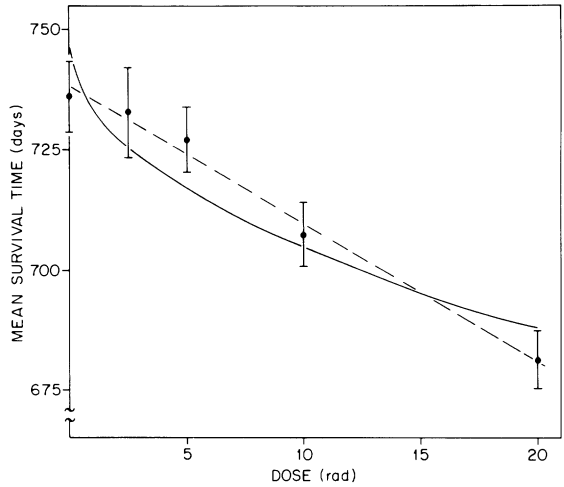

FIG. 3. Pooled data on mean survival times (with standard errors) for mice exposed to fission neutrons from the HPRR. Solid line is the least-squares fit for the square root of dose model. Dashed line is for the linear model.

dose range if the slopes for both radiations are truly linear. We cannot rule out, however, the possibility of nonlinearity for neutrons (or γ rays), and the RBE at very low doses could be significantly higher than 10.

DISCUSSION

There are no a priori reasons for assuming that the dose–effect curve for life shortening from neutron exposures must vary as either a linear or square root function

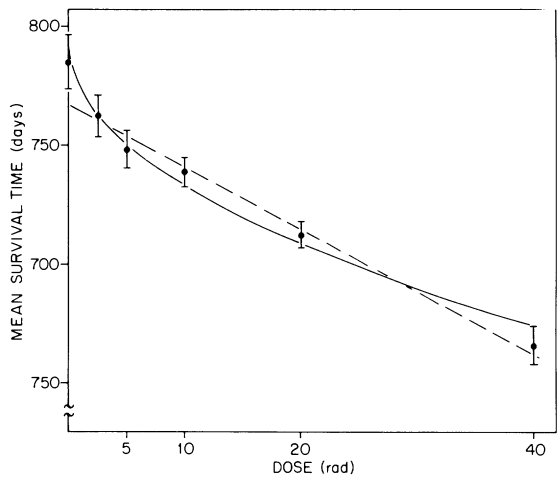

FIG. 4. Pooled data on mean survival time (with standard errors) for mice exposed to ^{252}Cf neutrons at various dose rates. Solid line is the least-squares fit to the square root of dose model. Dashed line is for the linear model.

of dose. When wide dosage ranges are considered, however, it is apparent that the curves are convex (decreasing slope with increasing dose) and an evaluation of these two alternative models in the low-dose (50 rad or less) range would seem reasonable for attempting to choose between potential extremes in models. However, if, as is now widely believed, the initial slope for many effects of low-LET radiation is approximately linear (*7*), the dual radiation action theory (*12*) would suggest that neutron effects should go with the square root of the dose to fulfill the prediction that the RBE of neutrons will vary inversely with the square root of neutron dose. An evaluation of data for a variety of responses has indicated that regardless of the shape of the curve for low-LET radiation the prediction of this systematic change in RBE seems to hold (*12*). Thus, to an extent, if the dose–effect curve for either low-LET radiation or neutron radiation is known, the other can be predicted.

In the present study we were unable to reject either the linear model or the square root of dose model in the dosage range of 40 rad or less. This was true even when the data for protracted exposures or fractionated exposures were pooled to improve the precision of the estimates of mean survival times. With high-dose-rate exposures delivered as single exposures or exposures to two equal fractions delivered at 1- or 30-day intervals we could not reject either model in the 0 to 50 rad range except for the case of the 30-day split where the square root of the dose model could be rejected but the linear model gave a reasonably good fit. In the dose range of 0 to 200 rad both models could be rejected although a more extreme curvature represented by dose to the 0.4 power gave an acceptable fit to the data for single exposures. No simple power function of dose gave an acceptable fit to the fractionated exposures in this extended dose range. A second-degree polynomial gave a satisfactory fit in all cases. When fitted to survival times the coefficient of the first-power term was negative and that of the second-power term was positive.

We conclude therefore that we were unable to distinguish unequivocally between the alternative models on an empirical basis. It is of interest to note, however, that in the three cases in which one model could be rejected in the low-dose range it has been the square root of the dose model that has been rejected (one case in the present study and two cases in the earlier study).

Kellerer and Rossi (*12*) have pointed out that a convex dose–response curve "could lead to an increased effect if a dose is split into two parts separated by a time interval that is long enough that the effects of the two doses are independent and simply additive." At high total doses where there is no question that the curve is convex, an increased effectiveness of long-protracted (*5*) or fractionated (*6*) exposures has been reported. An increased effectiveness of 30-day fractionated exposures at doses of 50 or 200 rad was also found in the present study. We did not find a significantly increased effectiveness after protraction or fractionation at doses of less than 50 rad.

If, as appears to be the case, a 30-day interval between doses is sufficiently long that the effects on survival time are independent and additive, then we may calculate the expected effects under the alternative models based on the data for single exposures. For single, high-dose-rate exposures in the range of 0–20 rad, the best estimates of mean survival times as a function of dose for the two models are

$$\text{Linear model} \qquad y = 733.8 - 2.98X,$$

$$\text{Square root model} \qquad y = 738.9 - 13.06(X^{0.5}),$$

it follows that the best estimates of life shortening (LS) with dose for single exposures are

$$\text{Linear model} \qquad LS = 2.98X,$$

$$\text{Square root model} \qquad LS = 13.06(X^{0.5}).$$

Using these equations we calculated the expected life shortening per fraction and the expected total life shortening for the animals given two exposures 30 days apart. From expected life shortening we calculated the expected mean survival times. These values, along with the observed values, are shown in Table IV. It is clear from this table that the linear model predicted the observed results more closely than did the square root model. In Fig. 5 we have plotted the observed mean survival times along with their 80% confidence intervals. The predicted results under the two models are also shown. On the basis of these findings we are inclined to reject the square root model in favor of the linear model for the dose range below about 20 to 40 rad. At higher doses there seems little doubt that the curve bends.

It may not be coincidental that this postulated critical dose range (20 to 40 rad) falls in the region where Rossi (*13*) has indicated an important transition occurs in the microdosimetry. According to Rossi, for neutrons in the energy range of 0.1 and 1 MeV, the number of cellular nuclei of higher organisms that are traversed by one neutron secondary is proportional to dose at doses substantially less than 25 rad. The dose to each traversed nucleus is on the order of 25 rad. Thus, in the low-dose range "the frequency of any cellular radiation effects must also be proportional to dose provided there is no interaction between irradiated cells." In this energy range and at a dose of about 25 rad all the nuclei will, on the average, be traversed by one neutron secondary. Our neutron sources gave a broad spectrum of energies, some of which were higher than 1 MeV. For the exposure configuration we used at the HPRR the median energy was 0.78 MeV and the mean was 1.28 MeV (*14*). The dose per traversed nucleus, however, drops slowly with increased neutron energies. For example,

TABLE IV

Expected Values Estimated from Alternative Dose–Response Models for Life Shortening and Mean Survival Times in Mice Receiving Two Equal Neutron Doses Separated by 30 Days

		Square root model			*Linear model*			
		Expected LS[a]			*Expected LS*			
Dose per fraction (rad)	*Total dose (rad)*	*per fraction*	*Total*	*Expected MST*[b]	*per fraction*	*Total*	*Expected MST*	*Observed MST ± SE*[c]
0	0	0	0	738.9	0	0	733.8	736.3 ± 7.38
1.25	2.5	14.6	29.2	709.7	3.7	7.4	726.4	738.8 ± 11.42
2.5	5	20.6	41.2	697.7	7.5	15.0	718.8	739.1 ± 11.29
5	10	29.2	58.4	680.5	14.9	29.8	704.0	719.3 ± 11.70
10	20	41.3	82.6	656.3	29.8	59.6	674.2	672.5 ± 11.64

[a] Life shortening (days).
[b] Mean survival time (days).
[c] Standard error.

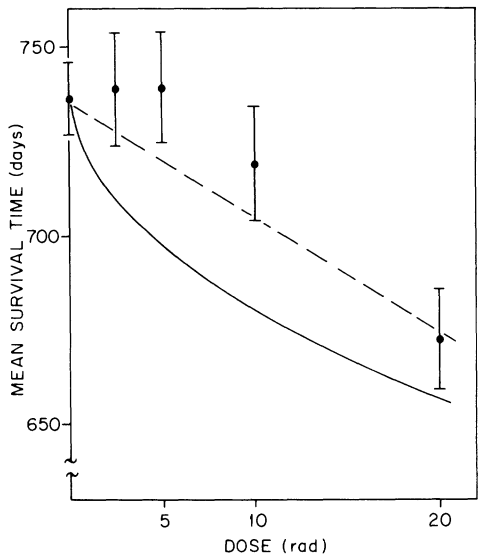

FIG. 5. Observed mean survival time (with 80% confidence intervals) for mice exposed to two equal doses of fission neutrons separated by a 30-day interval. Doses shown are total doses. The dashed line is for the expected survival times based on a linear fit to single dose data. The solid line shows expected survival times for the square root of dose model (see text).

increasing the neutron energy from 0.43 to 5.7 MeV decreases the dose per nucleus traversed by only a factor of two (*12*). For these reasons we conclude that, on the average, the dose per traversed nucleus was around 25 rad in this study.

Life shortening in irradiated mice has been reported to result primarily from an increased incidence and/or earlier onset of malignant neoplasms particularly in the low to moderate dose range (*15, 16*). A preliminary evaluation of some of the data from the present study supports this conclusion. It follows from microdosimetric considerations that further studies of neutron effects in the dose range of 0 to 50 rad should provide insights into mechanisms of radiation carcinogenesis. For example, if it can be shown that the dose–effect relationship is linear at doses of 20 rad or less but curvilinear at higher doses, can it be concluded that there is no interaction between cells whose nuclei are traversed by one neutron secondary but that interactions begin to occur when a significant number of cells are traversed by two or more secondaries? It would be important also to examine dose–effect curves and fractionation effects for tumors in tissues that do not normally show cell turnover and which do not respond to radiation (cell killing) by undergoing proliferation. These effects should be contrasted to those seen in proliferating tissues.

It may be possible to determine empirically whether, in the range of 0 to 20 rad, the curve for life shortening is a linear or a square root function of dose by increasing the precision of the survival estimates through increased sample sizes. Rather large samples would be needed, however. A more economical and perhaps more rewarding approach would be to use more fractionation schedules with doses per fraction of less than 20 rad but with total doses of 50 rad or more. If linearity is obtained with

the fractionated exposures then the presumption is strong that the effects are linear with dose in the low-dose range. Further, with this approach additional information on interaction times could be obtained.

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