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RADIOBIOLOGY OF LARGE ANIMALS

David C. L. Jones, et al

Stanford Research Institute

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RADIOBIOLOGY OF LARGE ANIMALS

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13 ABSTRACT

A review has been completed of all experiments concerned with radiation-induced lethality in sheep and conducted by the Naval Radiological Defense Laboratory and/or the Stanford Research Institute under the auspices of the Defense Civil Preparedness Agency. After consideration of a series of experiments involving sheep exposed at various continuous, intermittent, or combined irradiation schedules at does rates of 4 R/hr or less, it appears that the major factor determining mortality in such experiments is the average dose per day over the entire exposure period, and not the specific dose rate during the irradiation itself. A mathematical model has been developed that describes the dynamics of injury and repair as a function of daily average dose rate. Studies of cell kinetics in the bone marrow of mice have revealed that differences in mortality following irradiation at different dose rates may be related to corresponding differences in repopulation rates in this critical time, rather than with differences in cell destruction at the two dose rates.

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RADIOBIOLOGY OF LARGE ANIMALS

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SUMMARY

The Problem

In the event of a nuclear detonation, human populations will be exposed to ionizing radiations. A significant component of the radiation hazard will be due to fallout, which produces a wide range of radiation dose rates. The extent, and sometimes the variety, of radiation response depends on both the dose received and the dose rate at which it is delivered. Studies of the interrelationships among dose, dose rate, and response in a variety of animal species, particularly those approximating human size, are the primary source of information on which to base an evaluation of the potential radiation hazard to human populations.

The Findings

Review and summary of all studies to date concerned with radiation lethality in sheep have been completed. Lethality at high (30 to 660 R/hr) dose rates is adequately described by a linear relationship between exposure dose rate and $LD_{50/60}$. At low exposure dose rates ($\leqq$ 4 R/hr), however, the $LD_{50/60}$ depends more on the average daily dose than on the actual dose rate during irradiation, regardless of whether the irradiation is continuous, intermittent, or a combination of more than one exposure dose rate. Within limits, the relationship between $LD_{50/60}$ and average daily exposure can be adequately estimated from a mathematical model in which the rate of repair is an exponential function of the average daily exposure.

The pattern of recovery following radiation exposure depends on the level of net injury at the end of the exposure period. If the net injury at this time is not potentially lethal (within 60 days), the sheep shows a pattern of further decreasing radioresistance during the first week post-exposure, a transient period of increased radioresistance (even to supranormal levels) during the second week, then enters a second period of decreased radioresistance, which may persist for as long as several months. Because studies of radioresistance after potentially lethal doses of radiation are somewhat impractical from an economic/operational

viewpoint, the pattern of post-exposure radioresistance has not been determined, but a transient period of supranormal radioresistance does not appear likely. Decreased radioresistance in sheep surviving doses in the lethal range has been shown to persist for at least several months post-exposure.

Previous studies in mice had shown that, although the $LD_{50/30}$ at 1800 R/hr (868 R) was about two-thirds that at 200 R/hr (1359 R), the survival of bone-marrow colony forming units (CFU) was about the same at the two markedly different dose rates, indicating that the differential lethality did not appear related to the number of surviving stem cells. From recent experiments in mice exposed to 750 R at 1550 or 155 R/hr, it appears that the repopulation rate of the bone-marrow cells after the higher dose-rate exposure is appreciably less than after the lower dose-rate irradiation on the basis of both CFU count and on total marrow cellularity estimation.

ADMINISTRATIVE INFORMATION

The purpose of this Task Order is to continue the research previously conducted under Work Unit No. 2531D of Work Order DAHC20-67-C-0149 at the U.S. Naval Radiological Defense Laboratory. In August 1969, following the Defense Department's decision to close NRDL, the Defense Civil Preparedness Agency (then the Office of Civil Defense) was awarded Stanford Research Institute Contract No. DAHC20-70-C-0219 to continue this work (and three related work units). The Objective and Scope of Work as given in the pertinent Research Task Order attached to that contract are as follows:

> "OBJECTIVE: To improve quantitative models of radiation injury and recovery applicable to man through determination of the effects of protracted gamma irradiation on appropriate mammalian species of large animals in terms of radiation injury, recovery, and physiological alterations."
>
> "SCOPE OF WORK: To evaluate the hazards of nuclear warfare insofar as human population is concerned, it is essential to know more about the biological effects of the protracted irradiation characteristic of exposure in a fallout field. Since in fallout fields the dose rate will range from less than one rad per hour to several hundred rads per hour, our prediction capability must be extended to include the lower dose rate in particular. Information derived from chronic irradiation studies with large domestic animals whose radiation sensitivities and recovery processes are more comparable to those of man would be particularly pertinent."

Based on the Task Order quoted above, a Work Plan (dated August 1, 1971) was prepared, containing a list of milestones and an anticipated schedule for completion of these. This Work Plan was approved by the cognizant representatives of the Defense Civil Preparedness Agency.

With the publication and distribution of this report, all contractual requirements have been satisfied. In every respect, the objectives and scope of the contracted work, as further detailed in the approved Work Plan, have been successfully accomplished and completed on schedule.

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I INTRODUCTION

This is the third annual report on this project, which was designed to provide information on which to base estimates of the potential radiation hazards to humans consequent to fallout from nuclear detonations. The major thrust of this program is toward the development of models for estimation of hazards from low dose-rate gamma irradiation by establishing its effects on large animals, particularly sheep, in terms of injury accumulation and recovery.

The following areas of investigation were covered during the current contract year:

(1) A study of the $LD_{50/60}$ of sheep exposed to radiation doses of 280 R given at 3.5 R/hr at intervals of one or two weeks.

(2) A study of the total bone-marrow cell count in sheep given several radiation treatments.

(3) A study of the return of bone-marrow total cell count and colony-forming cell count in mice after irradiation at different dose rates to investigate the role of cell count replacement in determining the value of the LD_{50}.

(4) A study of the $LD_{50/60}$ of sheep given weekly radiation exposure cycles as follows: 140 R given at 3.5 R/hr, followed each week by either (a) 100 R given at 0.9 R/hr, or (b) 50 R given at 0.45 R/hr., or (c) no irradiation.

(5) Review and summary of all of the sheep experiments performed under Defense Civil Protection Agency (DCPA) support, both at the Naval Radiological Defense Laboratory and at Stanford Research Institute.

(6) Formulation of a mathematical model for sheep exposed to chronic or continuous low dose-rate irradiation that relates rate of recovery from radiation injury to the rate of exposure to the radiation and gives a method for calculation of the $LD_{50/60}$ at a given exposure rate.

Studies (1) and (2) cited above have been completed and reported in detail previously (1). Results of studies (3) and (4) are reported in Section II of this report. The review and summary of sheep experiments is presented in Section III of this report, and the mathematical model for recovery rate and exposure rate is presented in Section IV.

II FOURTH-QUARTER PROGRESS

Radiation Injury in Sheep Exposed to Varying Dose Rates

A series of experiments were done in the Spring of 1972 in which sheep were exposed to ^{60}Co gamma radiation in the following patterns:

(1) Exposure to 3.4 R/hr (midline-air) for 41 hr (total dose, 140 R), followed by exposure to 0.9 R/hr for 111 hr (total dose, 100 R). The cycle was repeated each week for totals of two, three, four, and five exposure cycles for different groups (Experiment 19).

(2) Exposure to 3.4 R/hr for 41 hr (total dose, 140 R), followed by exposure to 0.45 R/hr for 111 hr (total dose, 50 R). The cycle was repeated each week for totals of three, four, five, and six exposure cycles for different dose groups (Experiment 20).

(3) Exposure to 3.4 R/hr for 41 hr (total dose 140 R), followed by removal from the radiation range for the remainder of the week. The cycle was repeated each week for totals of six, seven, eight, and nine exposure cycles for different dose groups (Experiment 18).

The purpose of the experimental series was to investigate the effects of cyclic low dose-rate irradiation in experiments analogous to a post-attack situation in which men might work and be exposed to radiation at one dose rate, and periodically retire to shelters having various protection factor efficiencies for rest and reduction of the overall exposure rate.

The 60-day mortality results of Experiment 19 (140 R at 3.4 R/hr and 100 R at 0.9 R/hr; weekly dose, 240 R) are summarized in Table 1. The estimated $LD_{50/60}$ was 680 R, with confidence limits of 513 to 817 R.

Table 1

SIXTY-DAY MORTALITY IN SHEEP FOLLOWING REPEATED WEEKLY EXPOSURE TO COBALT-60 GAMMA RAYS ON THE FOLLOWING SCHEDULE: 3.4 R/HR (MIDLINE-AIR) FOR 41 HOURS (TOTAL DOSE=140 R), FOLLOWED BY 0.9 R/HR FOR 111 HOURS (TOTAL DOSE=100 R; TOTAL WEEKLY DOSE=240 R)
(Experiment 19)

Total Dose (R)	Mortality Freq.	Mortality %	Survival Time (Days)
483	3/15	20	7, 12, 42
725	11/15	73	6, 6, 8, 9, 12, 14, 2 x 15, 16, 18, 24
954	9/15	60	9, 10, 12, 15, 2 x 17, 22, 24, 34
1192	13/15	87	-8, -4, -1, 0, 1, 3, 4, 5, 6, 8, 14, 23, 29

Estimated $LD_{50/60}$: 680 R
95% Confidence Limit: 543-817 R
Mean Survival Time ± Standard Deviation: 12.3 ± 10.4 days

Estimated $ID_{50/60}$ without second dose group: 760 R
95% Confidence Limit: 600-921 R

As shown in the table, mortality results are not entirely satisfactory, in that the mortality was higher at 725 R than at 954 R. If the latter dose group is disregarded as an aberrant idiosyncrasy and the $LD_{50/60}$ is recalculated, the value is 760 R, a difference of 12%. Regardless of which value is used, however, it is apparent that with this alternating dose-rate schedule the $LD_{50/60}$ was reached in around 20 to 22 days.

The 60-day mortality results of Experiment 20 (140 R at 3.4 R/hr and 50 R at 0.45 R/hr; weekly dose, 190 R) are summarized in Table 2. The estimated $LD_{50/60}$ was 920 R, with confidence limits of 823 to 1018 R. The mortality results are reasonably satisfactory, as shown in the table. With an alternating dose-rate schedule of these parameters, the $LD_{50/60}$ was reached in about 34 days.

The 60-day mortality results of Experiment 18 (140 R at 3.4 R/hr, weekly increment) are shown in Table 3. The estimated $LD_{50/60}$ was 883 R, with confidence limits of 757 to 1009 R, and the $LD_{50/60}$ was reached in 37 to 44 days. Although the table does not show any serious distortions in dose-response relationships, the overall results appear somewhat unsatisfactory as far as $LD_{50/60}$ is concerned. First, the $LD_{50/60}$ for Experiment 18 is lower than that for Experiment 20, even though the exposure sequence for Experiment 20 includes the extra 50 R/week given at 0.45 R/hr. Second, Experiment 18 is a repetition of Experiment 13, which was done a year ago. The $LD_{50/60}$ for Experiment 13 was 1016 R (2), a value clearly greater than that for Experiment 18. The $LD_{50/60}$ for Experiment 13 is, in turn, larger than for Experiment 20, which is what would be expected with the extra 50 R/week given in Experiment 20. Moreover, an experiment somewhat related in design to Experiments 13 and 18, in which the doses were 280 R and the intervals were 2 weeks (Experiment 16) provided an $LD_{50/60}$ of 1059 R, in agreement with Experiment 13.

It is concluded that the results of Experiment 18, although not necessarily erroneous, appear aberrant to the extent that they do not fit with the results of the other experiments. For the moment, judgment

Table 2

SIXTY-DAY MORTALITY IN SHEEP FOLLOWING REPEATED WEEKLY EXPOSURE TO COBALT-60 GAMMA RAYS ON THE FOLLOWING SCHEDULE: 3.4 R/HR (MIDLINE-AIR) FOR 41 HOURS (TOTAL DOSE=140 R), FOLLOWED BY 0.45 R/HR FOR 111 HOURS (TOTAL DOSE=50 R; TOTAL WEEKLY DOSE=190 R) (Experiment 20)

Total Dose (R)	Mortality Freq.	Mortality %	Survival Time (Days)
568	1/15	7	9
757	4/15	27	2, 8, 9, 14
954	6/15	40	-7, 7, 18, 29, 31, 39
1145	13/15	87	0, 6, 7, 10, 2 x 18, 20, 21, 22, 23, 29, 30, 33

Estimated $LD_{50/60}$: 920 R
95% Confidence Limit: 823-1018 R
Mean Survival Time ± Standard Deviation: 16.5 ± 11.7 days

Table 3

SIXTY-DAY MORTALITY IN SHEEP FOLLOWING EXPOSURE
TO COBALT-60 GAMMA RAYS AT 3.4 R/HR (MIDLINE-AIR) FOR
41 HOURS ONCE EACH WEEK (WEEKLY DOSE=140 R)
(Experiment 18)

Total Dose (R)	Mortality Freq.	Mortality %	Survival Time (Days)
840	4/9	44	11, 16, 28, 32
980	5/9	56	11, 12, 16, 22, 50
1120	8/8	100	-4, -2, 0, 12, 14, 22, 25, 29
1260	8/9	89	0, 2, 7, 2 x 9, 12, 15, 20

Estimated $LD_{50/60}$: 883 R
95% Confidence Limit: 757-1009 R
Mean Survival Time ± Standard Deviation: 14.7 ± 12.2 days

is withheld on the significance of the results of Experiment 18. When an appropriate analysis can be made, Experiment 18 may make a useful contribution to the evaluation of the variability of the $LD_{50/60}$ results in general. The results of Experiments 19 and 20 have been incorporated in the detailed considerations presented in Section IV.

Bone-Marrow Total Cell Count and Colony-Forming Cell Count in Mice after Irradiation at Different Dose Rates

It was previously shown (3) that when mice were irradiated at two different dose rates for which the $LD_{50/30}$ was significantly different, the survival of colony-forming cells (CFU) in the bone marrow was the same for equal doses at the two rates up to a dose of 500 R. It was concluded that the difference in $LD_{50/30}$ at the two dose rates did not depend on differences in survival of bone marrow stem cells, and that the $LD_{50/30}$ in general did not depend exclusively on bone marrow stem-cell survival.

Studies during the current year have been conducted to determine whether the difference in $LD_{50/30}$ at different radiation dose rates might be caused by differences in rate of recovery of bone marrow stem-cell or total cell population in the first couple of weeks following irradiation. Two types of experiments were performed: (1) Donor mice were irradiated with 750 R of ^{60}Co gamma rays at either 1550 R/hr (1549.3 ± 23.5) or 155 R/hr (155.55 ± 2.35), and femurs of the donor mice were assayed for CFU by grafting cells into irradiated recipients at various times between 4 and 15 days after irradiation of the donors. (2) Mice were irradiated with 500 R of ^{60}Co gamma rays at either 1550 R/hr or 155 R/hr, and groups of mice were injected with ^{59}Fe and assayed for total marrow cell count at various times between 4 and 11 days after irradiation.

The results of the first type of experiment, the CFU assay, are summarized in Table 4. The values for CFU/femur at day 0 after irradiation were estimated by extrapolation of the survival curves of the CFU; the remaining values were measured directly. It can be seen that at

Table 4

NUMBER OF COLONY-FORMING CELLS PER FEMUR IN MICE
AFTER IRRADIATION WITH 750 R OF ^{60}Co GAMMA RAYS AT 1550 R/HR OR 155 R/HR

Day After Irradiation	Mean CFU per Femur 1550 R/HR	Mean CFU per Femur 155 R/HR
0	1.03	1.26
4	4.75	9.58
6	3.75	40.4
8	18.27	323.2
10	108.0	217.8
12	233.9	624
15	362.5	1473

Normal CFU/femur, unirradiated mouse: 2200 every time except the day of irradiation the number of CFU/femur for the mice irradiated at 155 R/hr was greater than that for mice irradiated at 1550 R/hr. The ratio of counts of CFU/femur for the two dose rates ranged from 2 to 20.

It was originally intended that the data on CFU replacement in the bone marrow be analyzed in terms of a general rate function and a cell doubling time. Although it is clear from the data shown in Table 4 that the rate of replacement of CFU is greater for the 155 R/hr irradiation than for 1550 R/hr, the actual regrowth process appears to be a complex series of events not amenable to general rate function analysis.

The results of the second type of experiment, the total bone-marrow cell count assay, are summarized in Table 5. After irradiation at 1550 R/hr, the cell count was 19% of control values at day 4, 72% of control at day 7, 59% at day 8, and 176% at day 11. After irradiation at 155 R/hr, the cell count was 58% of control values at day 4, 88% at day 11. Thus, at all time points measured, the total cell count after irradiation at 155 R/hr was greater than the total count after 1550 R/hr, although not all of the differences are significant.

An interesting possibility is suggested by the cell count assay at four days after irradiation. The mean total cell count after 1550 R/hr was very much below that after 155 R/hr. It may be that the dose-survival curves for stem cells (CFU) are independent of dose rate, but the dose-survival curves for more differentiated cells are not. Increased survival of intermediate cells in the bone marrow production line could lead to a sparing action on the stem cells, allowing a more rapid replacement of the stem cells and a greater resistance to the lethal effects of the radiation in the whole animal. This possibility merits some further investigation and will be explored during the next contract year.

Table 5

TOTAL NUMBER OF BONE MARROW CELLS IN MICE AFTER IRRADIATION WITH 500 R OF ^{60}Co GAMMA RAYS AT 1550 R/HR OR 155 R/HR

Day After	1550 R/hr		155 R/hr	
Irradiation	Cells x 10^{-8}	90% c.i.	Cells x 10^{-8}	90% c.i.
4	2.16	0.94-3.38	6.56	4.17- 8.97
7	8.20	5.13-11.29	9.94	7.53-12.61
8	6.68	6.03-7.34	13.20	9.33-17.05
11	19.85	14.78-24.88	21.20	15.66-26.79

Normal total number of cells, unirradiated mouse,
11.30 x 10^8 (10.07-12.55)

All values adjusted to a mean mouse weight of 22.5 g.

III SUMMARY OF NRDL AND SRI STUDIES TO DATE

In summarizing the studies using sheep as an experimental animal and performed by NRDL and SRI under the auspices of the DCPA, we have chosen to categorize them according to type of experimental procedure and concept of the study. In som cases, an experiment may have been originally planned or presented from a different viewpoint. There have been previous, more detailed summaries including some of the experiments (4-7). The categories for the present summary are:

(1) High dose-rate radiation lethality
(2) Low dose-rate radiation lethality
(3) Injury accumulation during low dose-rate irradiation
(4) Continuous exposure to death at low dose rates
(5) Periodic low dose-rate irradiation
(6) Post-exposure radiosensitivity

The subsequent section, concerned with the mathematical treatment of low dose-rate lethality data for sheep, provides a logical extension of this summary and a synthesis of most of the results. Hematologic measurements have been reported in detail elsewhere for those experiments done before the last quarter of the current year (1-3, 8-12). A summary of all hematologic findings for the sheep will be attempted during the next contract year.

High Dose-Rate radiation Lethality

High dose-rate exposure is used here to describe a single continuous exposure where a predetermined dose is delivered over a period ranging from a few minutes to a few hours, and where mortality occurs within a few days to a few months after exposure and is thought to be related to gastrointestinal and/or hematopoietic injury. This type of exposure has been traditionally considered analogous to "instant damage," and not involving concepts of rate of accumulation of injury or recovery during the exposure. From the data summarized here, this latter viewpoint appears open to question.

There have been ten experiments involving high dose-rate exposure of California-bred wethers to either ^{60}Co gamma rays or to 1 MVP X-rays. The results are summarized in Table 6. Six of these studies were done at NRDL, one at AWFL, and three at SRI. During an earlier detailed consideration of these data (5), results for Study 4 (Table 6) were not available, and the data were considered to be best represented by the linear regression equation (correlation coefficient = -0.82) $Y = 356 - 0.156X$, where Y is the $LD_{50/60}$ in R and X is the dose rate in R/hr. Subsequent inclusion of the Study 4 data results in a minor change in the calculated regression equation ($Y = 354 - 0.143X$, correlation coefficient = -0.77). In either interpretation, the following points appear to summarize adequately the relationship between dose rate and lethality for sheep exposed to a high dose rate: The $LD_{50/60}$ appears to vary with dose rate; the relationship appears reasonably described by a linear regression equation; and the relationship should be considered in evaluating the $LD_{50/60}$ at a high dose rate, especially when considering experimentally complex exposures where a high dose-rate value is used as a standard for evaluating injury accumulation or recovery during or after other types of exposures.

Low Dose-Rate Lethality

Four studies have been conducted on sheep irradiated with ^{60}Co gamma rays in a single, continuous terminated exposure. The first two studies, at 3.6 and 2.0 R/hr, were done at NRDL, the last two, at 0.9 and 0.8 R/hr at SRI. The relationships among the results of these studies, which are summarized in Table 7, will be included in the more general discussion presented in Section IV. In brief, the $LD_{50/60}$ at 3.6 R/hr is about twice those observed in comparable high dose-rate exposures; it increases further as the dose rate decreases. There is no significant difference between the $LD_{50/60}$ at 0.9 and at 0.8 R/hr. It is of interest that the results of the latter two studies indicate that dose rates of about 1 R/hr are effective in inducing lethality within the conventional 60-day, post-irradiation period, even though a smaller dose delivered at a similar dose rate may not result in appreciable residual injury at the end of the exposure period, as discussed in the next part of this section.

Table 6

RELATIONSHIP BETWEEN $LD_{50/60}$ AND DOSE RATE FOR SHEEP EXPOSED AT HIGH DOSE RATES. DOSES AND DOSE RATES EXPRESSED AS MIDLINE-AIR. EXPOSURE BILATERAL OR QUADRILATERAL IN ALL CASES. RADIATION SOURCE WAS ^{60}Co UNLESS OTHERWISE SPECIFIED.

Study Number	Dose Rate (R/Hr)	No. of Animals	$LD_{50/60}$ (R)	95% Conf.Lim. Lower (R)	95% Conf.Lim. Upper (R)	References
1	660	112	237	215	257	6, 13-15
2	578	60	262	241	283	9, 16
3	573	60	258	232	284	9, 16, 17
4	561	60	302	263	340	16
5*	450	118	258	243	276	13, 18
6*	450	74	314	292	344	19, 20
7*	450	58	320	261	349	19
8	426	55	298	230	338	21
9	261	72	318	291	343	15
10	30	60	338	313	369	15

* 1 MVP X-Rays

Table 7

RELATIONSHIP BETWEEN $LD_{50/60}$ AND DOSE RATE FOR SHEEP EXPOSED AT LOW DOSE RATES TO A SINGLE TERMINATED DOSE (MIDLINE-AIR) OF ^{60}Co GAMMA RADIATION.

Study Number	Dose Rate (R/Hr)	No. of Animals	$LD_{50/60}$ (R)	95% Conf.Lim. Lower (R)	95% Conf.Lim. Upper (R)	References
1	3.6	80	495	450	588	15, 22
2	2.0	48	637	538	698	15
3	0.9	60	1251	1149	1354	2
4	0.8	60	1117	687	1547	9

Injury Accumulation During Low Dose-Rate Irradiation

The experimental design for this type of study involves an initial single continuous exposure to a predetermined dose, followed immediately (within a few hours) by high dose-rate exposure to a challenge dose. This design is used to estimate the equivalent high dose-rate injury accrual during the low dose-rate exposure. It is particularly useful when it is not feasible to conduct a single low dose-rate lethality experiment--e.g., when the low dose-rate exposure is likely to require continuous exposure over a period of a year or so to achieve lethality. Variations in initial dose and dose rate and in challenge dose rate provide a wide range of possible studies. Seven experiments of this type have been conducted by NRDL and one by SRI. The results are shown in Table 8. Residual injury is obtained by subtracting the high dose-rate $LD_{50/60}$ measured at the end of the low dose-rate exposure from the high dose-rate $LD_{50/60}$ observed with no previous exposure. Thus, the amount of residual injury depends on the choice of the reference $LD_{50/60}$. Table 8 includes estimates based on the authors' selected reference value, on the value computed from the formula based on the data of Table 6, and on the latter value adjusted to a standard dose-rate of 600 R/hr. The fractional residual injury is computed by dividing the low dose-rate dose by the adjusted residual injury, and is used here only as an index of the pattern of injury accumulation during low dose-rate exposure.

In terms of the kinetics of injury accumulation during low dose-rate exposure, only the 3.6 R/hr experiments offer enough data points to allow any assessment. The four values at this dose rate are consistent with the hypothesis that net injury accrues more rapidly during the earlier portions of irradiation at a low dose rate than during the later period. These data do not, however, appear to be amenable to any more quantitative conclusion. At lower dose rates, insufficient data exist for any interpretation of the pattern of injury accumulation during exposure. It should be noted, however, that the rate of injury accumulation during 165 R at 1.85 R/hr appears to be somewhat less than for a comparable dose delivered at 3.6 R/hr. From the results of the 0.95 and 0.50 R/hr experiments, it appears that no net injury accumulation occurs

Table 8

NET ACCUMULATED INJURY IN SHEEP DURING LOW DOSE-RATE (LDR) EXPOSURE TO COBALT-60 GAMMA RADIATION AS ESTIMATED FROM HIGH DOSE-RATE (HDR) TITRATION. ALL DOSES AND DOSE-RATES ARE MIDLINE-AIR.

Study Number	Initial LDR Exposure: Dose Rate (R/Hr)	Initial LDR Exposure: Dose (R)	No. of Animals	Challenge Exposure: Dose Rate (R/Hr)	Challenge Exposure: $LD_{50/60}$ (R)	95% Conf. Lim.: Lower (R)	95% Conf. Lim.: Upper (R)	Estimated High Dose-Rate Residual Injury: Author (R)	Computed* (R)	(@600R/Hr) (R)	Residual Injury	References
1	3.6	400	43	510	71	40	107	199	210	200	0.50	15
2	3.6	305	48	530	86	63	110	174	192	185	0.60	6, 15, 22
3	3.6	165	47	660	133	106	162	104	127	131	0.79	6, 14, 15, 22
4	3.6	75	32	510	248	201	326	22	33	31	0.41	15
5	1.85	165	43	660	162	141	182	75	98	101	0.61	6, 14, 22
6	0.95	165	58	552	279	244	323	-42	-4	-4	-0.02	14, 22
7	0.50	165	92	552	268	224	328	-31	7	7	0.04	14, 22
8	0.46	727	58	514	168	118	196	--	112	107	0.15	2

* Y = 351-0.143X (see text).

after 165 R at these dose rates. Although this may be the case, there is definitely injury accumulation at 0.45 R/hr when the exposure is extended to 727 R, and, as noted previously, lethality does occur at 0.8 and 0.9 R/hr when the exposure period is long enough. Still et al. (6) suggested that a previous exposure at a high dose rate prevented, at least partially, recovery during immediately subsequent low dose-rate irradiation. In addition to the two pertinent experiments carried out at NRDL, three experiments were done at SRI to investigate this suggestion further. The results of these five experiments are summarized in Table 9. In both studies where the initial irradiation was at a low dose rate (about 3.8 R/hr), recovery during exposure is indicated by the finding that the equivalent high dose-rate $LD_{50/60}$, estimated from the data of Table 1, falls outside the 95% confidence limits for the total $LD_{50/60}$ in the combined low and high dose-rate exposures (see Table 9, Studies 1 and 2). When the low dose-rate irradiation was preceded by a high dose-rate exposure and a challenge dose delivered at a high dose rate (Studies 3 and 4) or a challenge dose delivered at a low dose rate (Study 5), the estimated equivalent high dose-rate $LD_{50/60}$ fell within the 95% confidence limits for the combined exposures, indicating that recovery during the low dose-rate exposures was not significant. It should be noted that the amount of recovery available for modification during low dose-rate exposures of 134-171 R is probably small, however, and the quantitative pattern of recovery during low dose-rate exposure has not yet been established (i.e., linear, exponential, etc.). Finally, the above findings depend on the particular values selected for the equivalent high dose-rate $LD_{50/60}$.

On the basis of present evidence, it appears that net injury accumulation during low dose-rate exposure is reduced by a previous high dose-rate exposure, but the parameters of this effect have not yet been characterized sufficiently to make any more quantitative statement.

Table 9

RELATIONSHIP BETWEEN RECOVERY DURING LOW DOSE-RATE (LDR) IRRADIATION AND PREVIOUS HIGH DOSE-RATE (HDR) EXPOSURE. DOSES AND DOSE-RATES EXPRESSED AS MIDLINE-AIR.

Study Number	No. of Animals	Initial HDR Dose (R)	Chronic LDR Dose (R)	Challenge $LD_{50/60}$ (R)	Total Exposure $LD_{50/60}$ (R)	Total Exposure 95% Conf. Lim. (R)	Est. HDR $LD_{50/60}$ (R)	References
1	17	--	165	133	298	271 - 327	260	6, 14, 15, 22
2	60	--	134	164	298	281 - 316	274	16
3	60	9	134	135	278	259 - 297	271	9, 16
4	60	45	134	91	270	254 - 285	272	9, 16, 17
5	91	155	--	171	326	279 - 364	290	19

Continuous Exposure to Death at Low Dose Rates

There have been two studies of the effect of low dose-rate irradiation under the special case of continuous exposure to death. The first was done at NRDL and the other at SRI. The results of these studies are summarized in Table 10. In this experimental design, it is apparent that each animal accrues an additional dose after it already has received a lethal amount of radiation. At 3.6 R/hr (terminated exposure), the $LD_{50/60}$ has been reported to be 495 R, while at 2.0 R/hr it has been estimated to be 637 R (12, 22). With radiation to death at corresponding dose rates, the comparable values are 1955 and 2142 R, respectively (Table 10). The differences between the values provide crude estimates of the excess, or "wasted," radiation associated with the exposure-to-death type of study. In the present case, these values amount to 1644 and 1318 R at 3.8 and 2.0 R/hr, delivered over periods of 19 and 29 days, respectively. Thus, the effect of this excess irradiation appears to be a function of the dose rate. From inspection of the individual survival times shown in Table 10, it is evident that excess irradiation at 3.8 R/hr results in a marked compression of survival times. This compression, together with an average after-survival value of 19 days, resembles the mortality pattern typical of high dose-rate exposures at or near the 100% lethal dose. Excess irradiation at 2.0 R/hr, however, does not appear to reduce the wide range of survival times, and the average after-survival time of 29 days is more typical of doses in the low-lethal range for terminated exposures.

Periodic Low Dose-Rate Irradiation

In general, this type of experiment involves recurrent exposure to the same dose increment (at regular intervals), and is usually designed to explore the relationships among various "packages" of radiation exposure. Single-exposure data provide the references to which periodic data are compared. Six major experiments have been concerned with periodic exposure of sheep at low dose rates, all conducted at SRI. The results are summarized in Table 11. It should be noted that all six experiments were done at similar dose rates, and that, with

Table 10

MORTALITY IN SHEEP EXPOSED CONTINUOUSLY (~ 23 HRS/DAY)
TO COBALT 60 GAMMA RADIATION AT 2.0 R/HR (23) OR 3.8 R/HR (9) MIDLINE-AIR

	Exposure at 2.0 R/Hr			Exposure at 3.8 R/Hr		
Survival (Days)	No. of Decedents	Cumulative Decedents	Accrued Dose (R)	No. of Decedents	Cumulative Decedents	Accrued Dose (R)
22				4	4	1923
23				4	8	2010
24				4	12	2098
25	1	1	1150	4	16	2185
26				2	18	2272
27				4	22	2360
28				2	24	2447
29	2	3	1334			
31	1	4	1426			
33	1	5	1518			
34	2	7	1564			
36	1	8	1656			
38	1	9	1748			
39	1	10	1794			
40	1	11	1840			
42	1	12	1932			
43	1	13	1978			
46	1	14	2116			
48	1	15	2208			
49	2	17	2254			
52	2	19	2392			
54	3	22	2484			
58	1	23	2668			
60	1	24	2760			

Table 11

RADIATION LETHALITY FOLLOWING PERIODIC EXPOSURE OF SHEEP TO COBALT-60 GAMMA RADIATION. ALL DOSES AND DOSE-RATES ARE MIDLINE-AIR.

Study Number	Exposure Dose-Rate (R/Hr)	Increment Dose (R)	Increment Interval (Days)	No. of Animals	$LD_{50/60}$ (R)	95% Conf.Lim Lower (R)	95% Conf.Lim Upper (R)	References
1	3.7	19	1	36	1251	862	1639	10
2	3.4	140	7	36	1016	848	1184	2
3	3.4	140	7	35	883	757	1009	This rep. Sec.II
4	3.4	140	7*	60	920	823	1018	This rep. Sec.II
5	3.4	140	7**	60	680	543	817	This rep. Sec.II
6	3.5	280	14	36	1059	968	1151	1

* 0.45 R/Hr during interim.

** 0.9 R/Hr during interim.

two exceptions, the incremental doses and intervals were selected to provide a dose rate of about 20 r/day averaged over the entire exposure period. In two cases, animals were maintained in very low dose-rate exposure fields during the intervals between incremental exposures at 3.4 R/hr. The data from these studies are presented in this section largely for documentation purposes. Studies 3 through 5 have been discussed in some detail in Section II, and all of them are included in the analysis presented in Section IV. Another preliminary experiment involving daily exposure to 100 R at 500 R/hr has been previously reported (14), but has not been included here because of the high dose rate, and because the results were somewhat equivocal.

Post-Exposure Radiosensitivity

Experiments designed to evaluate residual injury, or its corollary, recovery, involve a challenge exposure given at some interval after an initial exposure. Although the initial irradiation may involve a variety of exposure regimes, the challenge exposure is usually at a high dose rate. According to the primary concept under consideration, the reference value for evaluating the response to the challenge exposure is either the $LD_{50/60}$ for previously unirradiated animals, or the challenge $LD_{50/60}$ as measured immediately after the end of the initial exposure.

There have been 21 studies of this type, all done at NRDL. An additional three studies, designed somewhat differently, have been done at SRI, which will be discussed later. Tables 12 and 13 summarize the available NRDL data for sheep initially irradiated at high and low dose rates, respectively. The values for the estimated $LD_{50/60}$ at the end of the initial sublethal exposures are found by subtraction from the values listed in Table 6 for high dose-rate initial exposure, or from the values listed in Table 7 for low dose-rate initial exposure. The recovery values shown in Tables 12 and 13 use the response to a challenge exposure at the end of the initial exposure as a reference point, since these data result from studies primarily concerned with the recovery

Table 12

POSTIRRADIATION RECOVERY IN SHEEP EXPOSED TO ^{60}Co GAMMA RAYS OR 1 MVP X-RAYS AT HIGH DOSE-RATES

Study Number	Initial Exposure Dose-Rate (R/Hr)	Initial Exposure Dose (R)	Initial Exposure $LD_{50/60}$ (R)	No. of Animals	Time Interval (Days)	Challenge Exposure Dose-Rate (R/Hr)	Challenge Exposure $LD_{50/60}$ (R)	Challenge Exposure 95% Conf. Lim Lower (R)	Challenge Exposure 95% Conf. Lim Upper (R)	Estimated Net Recovery (R)*	Estimated Net Recovery %*	References
1	660	165	72	48	7	660	86	67	106	14	8	6
2	450	177	75	47	11	450	111	74	141	36	20	6, 20
3	450	177	75	60	16	450	275	NG	NG	200	113	6, 20
4	450	177	75	59	20	450	324	271	399	249	141	6, 20
5	450	177	75	59	24	450	207	151	253	76	75	6, 20
6	450	177	75	43	30	450	179	150	203	104	59	6, 20
7	660	165	72	39	31	660	185	106	213	113	68	6, 13, 20
8	450	177	75	48	45	450	193	164	219	118	67	6, 13, 20
9	450	177	75	46	75	450	218	144	269	143	81	6, 20
10	450	100	216	60	7	450	256	218	295	79	40	6
11	450	100	216	72	16	450	567	505	687	471	351	20

* See text.

Table 13

POSTIRRADIATION RECOVERY IN SHEEP EXPOSED TO ^{60}Co GAMMA RAYS AT LOW DOSE-RATES

Study Number	Initial Exposure Dose-Rate (R/Hr)	Initial Exposure Dose (R)	Initial Exposure $LD_{50/60}$ (R)	No. of Animals	Time Interval (Days)	Challenge Exposure Dose-Rate (R/Hr)	Challenge Exposure $LD_{50/60}$ (R)	Challenge Exposure 95%Conf.Lim Lower (R)	Challenge Exposure 95%Conf.Lim Upper (R)	Estimated Net Recovery (R)*	Estimated Net Recovery %*	References
1	3.9	305	86	36	4	660	118	41	167	32	21	6
2	3.9	305	86	48	7	660	157	126	186	71	47	6
3	3.9	305	86	48	15	660	361	296	445	275	182	6
4	3.9	305	86	34	30	660	163	73	199	77	51	6
5	3.9	165	133	42	7	660	218	185	270	85	82	6, 14, 20
6	3.9	165	133	48	15	660	546	500	699	213	397	6, 20
7	3.9	165	133	45	27	660	210	187	236	77	74	6, 14, 20
8	1.85	165	162	77	15	660	342	303	382	180	240	6, 14, 20
9	1.85	165	162	19	27	660	225	NG	NG	63	24	6, 14, 20
10	0.5	165	268	65	31	660	245	214	284	-23	74	14, 22

* See text.

process itself, beginning with the end of the initial exposure. Recovery in R was calculated from the formula Recovery = $LD_{50/60t} - LD_{50/60o}$, and Percent Recovery = 100 $((LD_{50/60t} - LD_{50/60o})/(LD_{50/60n} - LD_{50/60o}))$, where the subscripts refer to the $LD_{50/60}$ for animals not previously irradiated (n), animals challenged immediately at the end of an initial exposure (o), or at some later time (t). Since various studies use different $LD_{50/60n}$ values, and since the possible recovery using the above formulas is markedly affected by the initial dose, it is useful for present purposes to relate radiation sensitivity to that of the animal with no previous irradiation. With this viewpoint, the emphasis is on a conceptualization useful for evaluating the response of previously irradiated animals to another exposure in terms of a comparison with "normal" controls, rather than on an evaluation of how they have changed since the initial exposure. Accordingly, the values for the studies listed in Tables 12 and 13 have been recalculated and summarized in Table 14. In recomputation, values of $LD_{50/60n}$ computed from the formula given earlier in this report (Y = 354 - 0.143X) have been used to minimize inter-report variability. Also, an additional study, done at SRI, has been included (to be described below). From Table 14, it becomes obvious that the radiosensitivity of sheep immediately following an initial exposure is a function of the initial dose. Further, if the dose rate and the dose during the initial exposure are low enough, no difference from control radiosensitivity may be detectable immediately following the initial dose (e.g., 165 R at 0.95 or 0.50 R/hr). The change in radiosensitivity with time following the initial exposure is easily discernible: an increased radiosensitivity during the first week or so, a transient period of decreased radiosensitivity during the third week, and a second sustained period of increased radiosensitivity extending at least into the third month after the initial exposure. It should be noted that the evidence for a period of excess radioresistance (above control) comes entirely from studies where the initial exposure was sublethal.

Although the phenomenon of transient increased radioresistance has been observed in several other species (24, 25), there is no evidence

Table 14

RELATIVE RADIORESISTANCE* OF SHEEP FOLLOWING EXPOSURE TO ^{60}Co GAMMA RAYS OR 1 MVP X-RAYS

Initial Exposure		Time of Challenge (Days After End of Initial Exposure)													
(R/Hr)	(R)	0	4	7	11	15	16	20	24	27	30	31	45	75	90
660	165	0.28		0.33								0.71			
450	177	0.26			0.38		0.95	1.12	0.71		0.62		0.67	0.75	
450	100	0.75		0.88			0.96								
3.9	305	0.33	0.45	0.60		1.39					0.63				
3.9	165	0.51		0.84		2.10				0.81					
1.85	165	0.62				1.32				0.87					
0.95	165	1.01													
0.84	747**	0.67										0.89			0.67
0.50	165	0.97													

* Relative Radioresistance= $1-[(LD_{50/60n} - LD_{50/60i})/(LD_{50/60n})]$, where subscript (n) refers to previously unirradiated animals and (i) refers to previously irradiated animals.

** Average of initial exposures ranging from 657 to 897R.

that a similar phenomenon may exist after initial doses in the lethal range. Indeed, it seems unlikely that excess radioresistance would occur after lethal-range irradiation, particularly when the exposure is at a high dose rate. With respect to the observation that no discernible change from control radiosensitivity was observed immediately after exposure to 165 R at 0.95 or 0.50 R/hr, it should be noted that when a larger dose was given at 0.84 R/hr, significantly reduced radiosensitivity was observed immediately following the initial exposure as well as three months later in survivors of the second irradiation.

IV MATHEMATICAL ANALYSIS OF THE LETHAL RESPONSE OF SHEEP TO CHRONIC EXPOSURE

As the rate of exposure to ionizing radiation decreases, the effectiveness of the radiation for causing death also decreases, and a larger dose of radiation is required to produce a given level of mortality. The reason usually given for this dose-rate effect is that the injury caused by the radiation is partly repaired during exposure to the radiation. This analysis attempts to relate the apparent rate of repair of the radiation injury to the rate of exposure to radiation in sheep receiving dose rates of 4 R/hr and less. In this analysis, rate of exposure to radiation and rate of repair of radiation injury are both expressed as R/unit time, usually R/day.

It was noted in Section III of this report that the $LD_{50/60}$ of sheep exposed at dose rates of between 30 and 660 T/hr was adequately described as a linear function of the dose rate. In the analysis that follows, this relationship between $LD_{50/60}$ and dose rate is assumed to be

$$LD_{50/60} = 356 - 0.156 \times \text{dose rate}. \quad (1)$$

However, below 30 R/hr, the linear formula does not correctly predict the $LD_{50/60}$. The theoretical maximum $LD_{50/60}$ predicted by the linear formula is 356 R, whereas inspection of Table 15 shows that at 3.6 R/hr the $LD_{50/60}$ was 495 R, and at lower dose rates was even greater. It is postulated for the present analysis that at some dose rate between 30 R/hr and 4 R/hr, an additional repair mechanism is activated in the sheep that acts to reduce the effectiveness of the radiation. The mathematical characterization of this additional process is the subject of this analysis.

The data for the analysis of this supplementary repair were taken from Tables 7 and 11 and from Reference 2. For the exposure at 0.46 R/hr (2), the $LD_{50/60}$ value was recalculated on the assumption that the mean daily exposure rate was exactly half the exposure rate at 0.92 R/hr. This recalculation gives only a minor (2%) change in the $LD_{50/60}$.

The analysis of the repair of radiation injury during exposure is shown in Table 15. The upper part of the table presents the experiments in which the exposures were continuous throughout day and night, except for periods of from 0.5 to 2 hr/day downtime for feeding and watering the animals and performing other range services. The exposure rate expressed in R/day allows for the downtime. The measured $LD_{50/60}$ in R at each exposure rate is shown in the third column. The fourth column, labeled "Theoretical $LD_{50/60}$," is the $LD_{50/60}$ that would have been expected on the basis of Eq. (1) if the supplementary repair had not been present. The fifth column, labeled "Repair," is the difference between measured and theoretical $LD_{50/60}$ (columns 3 and 4). The sixth column, labeled "Exposure Time" is the number of days required at the given daily rate to deliver the measured $LD_{50/60}$. The seventh and eighth columns, labeled "Repair Rate" are, respectively, the amount repaired per day of exposure and the percentage of the daily radiation exposure that is repaired. In summary, the exposure of 3.6 R/hr was 84.6 R/day, the measured $LD_{50/60}$ was 495 R, the theoretical $LD_{50/60}$ was 355.44 R (356 - 0.156 x 3.6), the repair was 139.56 R, or 24.19 R/day, which was 28.5% of the rate of exposure.

Columns 2, 7, and 8 of the Table 15 show that, as the daily exposure rate decreased, the absolute repair/day also decreased, but at a slower rate, so that the repair/day became a rapidly increasing proportion of the daily exposure rate. The overall effect suggests that (1) the daily repair rate has some maximum value, which is approached asymptotically as the daily rate of exposure increases; and (2) the repair rate approaches the exposure rate as the daily rate of exposure decreases, becoming equal to the exposure rate at zero exposure rate. This relationship can be approximated by assuming that the repair rate is an exponential function of the exposure rate of the form.

$$R = R_o(1 - e^{-kD}) \qquad (2)$$

where R is the rate of repair per day, R_o is the maximum repair per day, k is a constant, and D is the rate of exposure per day.

Table 15

$LD_{50/60}$ DATA IN SHEEP RECEIVING PROTRACTED EXPOSURES TO ^{60}Co IRRADIATION

Exposure Rate		$LD_{50/60}$		Repair	Exposure Time	Repair Rate	
(R/Hr)	(R/Day)	Measured (R)	Theoretical (R)	(R)	(Days)	(R/Day)	(%)
3.6	85.5	495	355.438	139.562	5.789	24.108	28.2
2.0	47.5	637	355.688	281.312	13.411	20.976	44.2
0.915	20.050	1251.546	355.857	895.689	62.421	14.349	71.6
0.871	19.906	1117.328	355.864	761.464	56.130	13.566	68.1
0.457	10.025	1747.009	355.928	1391.081	174.265	7.983	79.6
2.36	34.286	760.395	355.632	404.763	22.178	18.251	53.2
2.70	27.143	920.220	355.579	564.641	33.903	16.655	61.3
3.47	25.518	996.881	355.459	641.422	39.066	16.419	64.3
3.39	22.467	1015.997	355.471	660.526	45.222	14.606	65.0
3.47	22.160	1113.309	355.459	757.850	50.240	15.085	68.1
3.70	19.305	1250.787	355.435	895.352	64.791	13.819	71.6

The method of fitting the data of Table 15 to Eq. (2) is as follows: (1) a reasonable probable value of R_o is assumed (from the data of Table 15 it was assumed to be 25 R/day); (2) the measured rate of repair per day is subtracted from the assumed value of R_o; (3) a calculation is made of the linear regression of $\log_e (R_o - R)$ on the rate of exposure per day (this regression calculation customarily includes the value of zero R/day exposure, for which $(R_o - R) = R_o$. The computed slope of the regression equation is the estimated value of k.

From the first fit of the data to Eq. 2, a revised estimate is obtained for R_o, as follows: From the regression equation, the expected values of $\log_e (R_o - R)$ are computed for each of the measured rates of exposure per day. The values of [expected $(R_o - R)$ + measured (R)] are new estimates of the value of R_o; if the mean value of the new estimate of R_o is different from the one used previously, a new cycle of calculation is performed, starting with the new estimate of R_o. With repeated re-calculations, the estimated value of R_o converges on some final value, which becomes accepted for general use.

A preliminary calculation of the values shown in the upper part of Table 15 indicated that they could be fitted acceptably to Eq. (2) by the method described above. Before proceeding with the calculation, we evaluated other experiments that could contribute to the analysis of protracted radiation in sheep. These experiments were those listed in Table 11, involving exposure at a low dose rate, mainly 3.5 R/hr, but not continuously throughout the day and night or at the same dose rate. The first experiment (Experiment 8), involving exposure at 3.7 R/hr for 5-1/3 hr each day, is shown in the second half of Table 15 as a mean exposure rate of 19.3 R/day. Three other types of experiment were: (1) exposure to 140 R at 3.5 R/hr once each week (Experiments 13 and 18); (2) exposure to 280 R at 3.5 R/hr every two weeks, and (3) exposure as in (2), with some of the groups receiving a terminal dose of 140 R (Experiment 16). The actual exposure times, including source downtime, were 42 and 84 hr for the 140 and 280 R exposures, respectively, and the one- or two-week intervals were defined as the time from the beginning of one exposure to the beginning of the next exposure. The intent was to provide an average of 20 R/day, 140 R/week in the form of several alternate "packaging" arrangements. However, when the significance of the daily average exposure rate became manifest, the procedures for the experiment were reexamined, and it was then noted that the average daily exposure rates for these experiments was greater than 20 R per day. For instance, if the 140 R exposure required 1.75 days to deliver, then two exposures at an interval of one week would result in 280 R being given in 8.75 days, for an average daily exposure rate of 32 R/day; three exposures at intervals of one week would give 420 R in 15.75 days, or 26.67 R/day, and so forth. As the total number of exposures increased, the average exposure rate approached 20 R/day, but within the limits of the $LD_{50/60}$ the average exposure rate was significantly greater than 20 R/day. It is assumed in the present analysis, and appears reasonably supported by the data, that effects of "dose packaging" intended in the original design turn out to be effects of "mean daily rate of exposure."

For analysis of the experimental data, the mean daily exposure rate was computed for each dose group in each experiment, and the mean daily exposure rate for the experiment as a whole was computed as the mean exposure rate for those groups showing partial mortality response. In the final analysis of the data, the following modifications were made:

(1) Experiment 18 was eliminated from the set because the $LD_{50/60}$ was abnormally low.

(2) Experiment 13 appears in the table under the exposure rate of 22.467 R/day and the $LD_{50/60}$ of 1015.997 R.

(3) Experiment 16 consisted of a series of exposures to doses of 280 R every two weeks at 3.5 R/hr. In the experimental design, some of the groups were given 140 R as the final exposure to make the group dose interval 140 R. For those groups receiving 140 R as a final dose--e.g., 2 x 280 + 140, 3 x 280 + 140, 4 x 280 + 140--the total dose received was greater than groups that terminated at 280 R--e.g., 2 x 280, 3 x 280, 4 x 280--

but the mean rate of daily exposure was less. When the groups receiving the two different terminal irradiations were separated and treated as independent experiments, the set of groups with the 140 R terminal irradiation had a mean exposure rate of 22.100 R/day and an LD_{50} of 1113.309 R, and the set of groups without the 140 R terminal exposure had a mean exposure rate of 25.518 R/day and an LD_{50} of 996.881 R. These two sets appear as separate entries in the lower half of Table 15.

(4) The last two experiments involved exposure of sheep to alternating dose rates on an around-the-clock basis (Experiments 19 and 20). The weekly dose for Experiment 19 was 140 R at 3.5 R/hr plus 100 R at 0.9 R/hr, for a total of 240 R/week, or 34.286 R/day. The $LD_{50/60}$ of 760.395 R for this experiment was calculated by elimination of group 2 of the experiment, which had excessive mortality. The weekly dose for Experiment 20 was 140 R at 3.5 R/hr plus 50 R at 0.45 R/hr for a total of 190 R/week, or 27.143 R/day. The $LD_{50/60}$ was 920.220 R.

The complete set of values in Table 15 was fitted to Eq. (2) by the procedure described above. The resulting regression line was

$$\log_e (24.898 - R) = 3.203 - 0.0399D \qquad (3)$$

and the exponential form

$$R = 24.898 (1 - e^{-0.0399D}) \qquad (4)$$

A plot of the resulting relationship between exposure rate and recovery rate is shown in Figure 1. The points in the figure are the experimental points listed in Table 15. The curved line is the fitted line corresponding to Eq. (4), and the horizontal line is the maximum repair rate asymptote of the fitted line. It can be seen that the fitted line is an excellent representation of the data points.

Backfitting of the fitted Eq. (3) was done to obtain the expected values of repair rate, $LD_{50/60}$, and time to $LD_{50/60}$ for the experimental points. Equation 3 predicts the rate of repair of radiation injury at

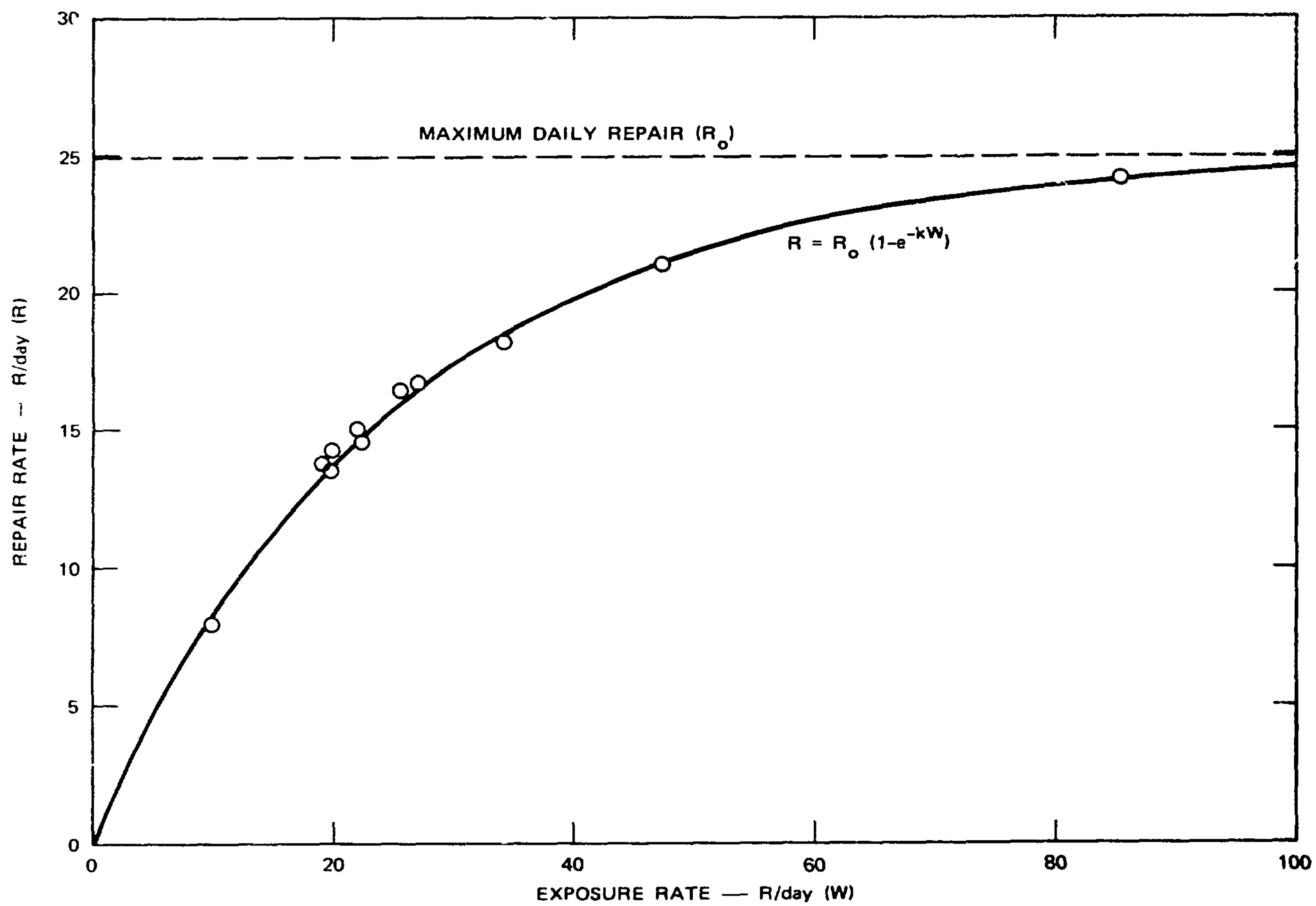

FIGURE 1 THE RATE OF REPAIR OF LETHAL GAMMA RADIATION INJURY IN SHEEP IN RELATION TO THE RATE OF RADIATION EXPOSURE

a given rate of radiation exposure. To calculate a predicted $LD_{50/60}$ for a given rate of exposure, the calculated rate of repair is subtracted from the rate of exposure, giving the net rate of accumulation of injury. The rate of accumulation of injury is divided into the theoretical $LD_{50/60}$ for the dose rate (Table 15, column 4), giving the calculated number of days required to reach the $LD_{50/60}$. The calculated number of days required to reach the $LD_{50/60}$, multiplied by the rate of exposure per day gives the calculated $LD_{50/60}$.

The measured and calculated values for repair rate, $LD_{50/60}$, and the time to reach $LD_{50/60}$ are compared in Table 16. Figure 2 shows the relationship between daily exposure rate and $LD_{50/60}$. The solid line in the figure is calculated from Eq. 3 with the further assumption that the radiation is given continuously throughout the day and night. The calculable differences between continuous and periodic exposure at the dose rates used experimentally are trivial in all cases. Figure 2 shows that, with the possible exception of the 10 R/day exposure, the agreement between calculated and measured $LD_{50/60}$ is excellent. At the 10 R/day exposure, the predicted rate of repair is in good agreement with the measured value (Figure 1, Table 16), but at this exposure rate the repair is about 80% of the dose, and small uncertainties in the rate of exposure or the rate of repair can make large contributions to the rate of accumulation of lethal injury.

The foregoing analysis shows that the data on $LD_{50/60}$ in relation to exposure rate in the sheep can be fairly represented by a model in which the rate of repair is assumed to be an exponential function of the exposure rate. Although the model gives excellent results, a further consideration of the limits and applications seems to be in order.

First, with respect to the model, it should be understood that the exponential model is used to generate a curve form with properties similar to the trend of the actual data. The fit between the model and the data is excellent--so good as to suggest that the dynamics of injury

Table 16

COMPARISON OF MEASURED AND CALCULATED VALUES FOR RECOVERY AND LD_{50} IN SHEEP RECEIVING PROTRACTED EXPOSURE TO ^{60}Co IRRADIATION

Exposure Rate (R/Day)	Repair Measured (R/Day)	Repair Calcd. (R/Day)	LD_{50} Measured (R)	LD_{50} Calcd. (R)	Time to LD_{50} Measured (Days)	Time to LD_{50} Calcd. (Days)
85.500	24.108	24.086	495	494.9	5.789	5.788
47.500	20.976	21.198	637	642.3	13.411	13.523
34.286	18.251	18.629	760.395	778.532	22.178	22.707
27.143	16.655	16.561	920.220	912.059	33.903	33.602
25.518	16.419	16.002	996.881	953.199	39.066	37.354
22.467	14.606	14.850	1015.997	1048.490	45.222	46.668
22.160	15.085	14.727	1113.309	1059.736	50.240	47.822
20.050	14.349	13.833	1251.546	1147.642	62.421	57.239
19.906	13.566	13.769	1117.328	1154.289	56.130	7.987
19.305	13.819	13.499	1250.787	1181.813	64.791	61.218
10.025	7.983	8.390	1747.009	2182.312	174.265	217.687

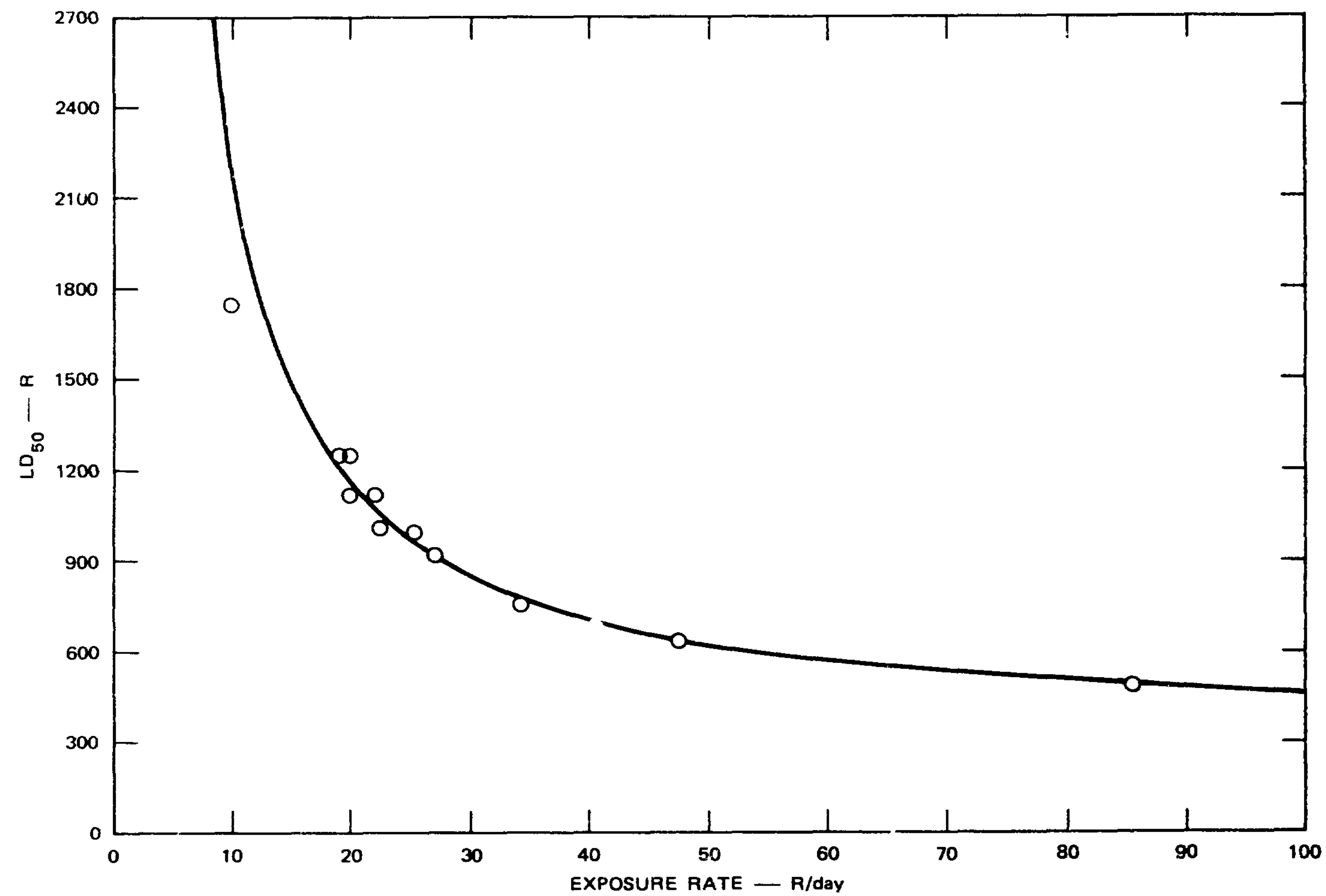

FIGURE 2 THE ACUTE LD_{50} IN SHEEP FROM GAMMA IRRADIATION IN RELATION TO THE RATE OF RADIATION EXPOSURE

repair do follow the mathematical model--but at present there is not a demonstrable biological mechanism to account for the form of recovery rate.

Second, as the model is constructed, it uses all $LD_{50/60}$ points with equal weight, irrespective of the confidence intervals of the individual measurements. The confidence interval of the exposure rate is also disregarded. In addition, because of the nature of the calculations, the uncertainty of the calculated rate of repair and the $LD_{50/60}$ partly depends on the rate of exposure. For all these reasons, calculation of reasonable confidence intervals for the regression line [Eq. (3)] and reasonable limits for the extrapolation of predicted $LD_{50/60}$ is a complex task that has been deferred until completion of the next series of experiments.

The model as it is constructed applies to sheep exposed at a low rate (~3.5 R/hr or less) continuously or at regular intervals spaced not more than two weeks apart. No attempt has been made to relate the data on repair rate by split dose studies (Tables 12 and 13) to the present model, and it appears problematic at present that such a relationship can be constructed in an analytic fashion. The effect of periodic exposure at high dose rates is currently unknown, although the evidence from one limited experiment (Experiment 14) indicates that repair of radiation injury does take place. The effect of periodic exposure at high dose rates is being investigated in the experimental program for the next contract year. Effects of combinations of high and low dose rate exposures are also not covered by the model, although some experiments have been done (Table 9). When the current studies of recovery during periodic high dose-rate exposures have been completed, the question of combinations of high and low dose-rate exposures will be reexamined. Finally, the model is applied to sheep with no previous radiation history. The evidence from reirradiation studies indicates that at three to six months after completion of chronic irradiation at low dose rates, there is a substantial amount of injury remaining in the animal, as judged by reduction of acute

$LD_{50/60}$ at high dose rates. The effect of previous irradiation exposure on the $LD_{50/60}$ at low dose rates is currently under investigation.

In addition to modifications of the present model to take into account the effects of high dose rate and high/low dose rate combinations, other possible modifications may be considered. The present model for repair of injury during irradiation assumes that the rate of repair of the injury does not become equal to the exposure rate until the exposure rate is zero. However, various experiments in mice suggest that there may be finite exposure rates at which the repair rate equals the exposure rate and no net accumulation of injury occurs. Suitable modifications of Eq. (2) can be considered to allow for the possibility that there is a cut-off exposure rate where accumulation of lethal injury does not occur.

Preliminary investigations have been made concerning the potential extension of the present mathematical model to other species. The literature contains extensive studies on protracted irradiation in the mouse and a few limited studies in the pig, dog, and burro. Unfortunately, most of the studies have involved irradiation of animals to death, with mean survival time of the animals used as the biological end point. The sheep is the only animal for which there are fairly extensive terminated exposure $LD_{50/60}$ data over a wide range of exposure rates, and all the data have been accumulated in this project. The task of extending the model will involve an attempt at conversion of the data on mean survival time into estimates of $LD_{50/60}$ for several dose rates and several animal species. This work will be undertaken during the next contract year.

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