species (*Rattus*) over a wide age and weight range. Additional material collected in the future for the same and different species might be expected to define this curve more sharply, but not to alter it in any essential manner. The two extremities, in particular, need further validation.

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Effect of X-Rays on Micronuclear Number in *Paramecium aurelia*[1]

R. P. Geckler[2] and R. F. Kimball

Biology Division, Oak Ridge National Laboratory, Oak Ridge, Tennessee

The reduced viability of exautogamous descendants of *Paramecium aurelia* exposed to x-rays or nitrogen mustard has been interpreted as the result of gene mutations or small chromosomal aberrations in the micronuclei (*1, 2*). In addition, other inherited changes may be involved (*2*). Among these, one that could be important is the loss of micronuclei immediately after irradiation. Although circumstantial evidence against such loss has been reported (*1*), no quantitative data on micronuclear frequency after exposure to mutagens have been available by which to test this hypothesis directly.

Experiments were therefore designed to evaluate the magnitude of variation of micronuclear number in relation to x-ray dosage to determine whether such variation could, in fact, contribute appreciably to the genetic results previously obtained with this organism.

Tube cultures of stock OR, variety 1 of *P. aurelia,* maintained by standard methods (*3*) were centrifuged and resuspended in fresh culture medium. One-ml samples of the resuspended cultures were exposed to x-rays in Lucite dishes on a rotating turntable. The x-ray source was a GE Maxitron operated at 250 kvp and 30 ma, with approximately 1 mm of Al inherent and no added filtration. The intensity measured by a nylon Victoreen chamber in air was 15–17 kr/min.

After exposure, samples of 15 animals were isolated individually from each group so that the effect on division and survival could be determined. The remaining animals were placed in test tubes with an excess of culture fluid and allowed to multiply for 24 hr at 27° C. At the end of this period, they were fixed in Schaudinn's fluid (60° C), stained by Dippell and Chao's (*3*) modification of the DeLamater technique, omitting the formalin mordant. The number of micronuclei per animal was then counted. The following types of animals were excluded from the count: animals containing more than two macronuclear fragments, animals partially hidden by other animals or debris, broken or fragmented animals, and animals that were not differentially stained. Dividing animals were accepted as single animals and the number of micronuclei taken to be half the total found.

Data from these studies are given in Table 1. It is of interest to note that there is a small variation in micronuclear number in unirradiated animals. In irradiated animals, the fraction with less than two micronuclei increased with increasing dose, whereas the fraction with two decreased. No unequivocal change occurred in the group with more than two micronuclei. By pooling the data from each exposure into groups of animals with (1) more than two micronuclei, (2) two micronuclei (the usual number), and (3) less than two micronuclei, the classes are large enough to test statistically. The data are heterogeneous, as shown by a χ^2 value of 112 for 12 degrees of freedom ($P < 0.01$).

The present study does not throw light on the mechanism by which abnormal numbers of micronuclei arise. However, the failure to produce more than about 10% of animals with less than two micronuclei

TABLE 1

EFFECTS OF X-RAYS ON MICRONUCLEAR DISTRIBUTION IN *Paramecium aurelia*

Dose (kr)	Survival in sample of 15 animals	Mean No. fissions of survivors (in 24 hr)	Percentage of animals with various numbers of micronuclei					Total No. animals counted
			4	3	2	1	0	
0	15	3.2	0	4	93	3	0.4	435
214.2	15	2.1	2	2	91	1	4	86
275.4	15	1.6	0	3	87	7	3	30
335.7	7	1.1	3	2	83	10	2	100
336.6	14	1.0	6	16	72	0	6	18
402.8	5	1.2	0	5	78	6	11	100
469.9	3	1.3	1	9	79	4	7	100*
537.4	2	0.5	0	7	57	19	17	83

* One animal exposed to this dose, but not included in the count, had 7 micronuclei.

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[2] Research and Medicine Division, AEC, Oak Ridge, Tenn.

except at doses above 400 kr is important, since it shows that loss of micronuclei is not a major reason for reduced vigor and death after autogamy. At 200 kr about 90% of the exautogamous clones die within a few divisions, and even at 10 kr about 80% are abnormal. The animals that lose micronuclei at these doses could account for only a small fraction of these effects. The known similarity in action between x-rays and nitrogen mustard makes it appear likely that similar results would be obtained with the latter.

These data also give evidence on the process of division delay in *Paramecium*. At the lower doses used, the division of the cytoplasm and the micronuclei must have been delayed to almost the same extent. Otherwise many more animals with abnormal numbers of micronuclei would have been found. At doses of a few kiloroentgens, it is known that there is practically no delay in cytoplasmic division (*4*). The possibility remained that micronuclear mitosis might show a major inhibition comparable to that shown by the grasshopper neuroblast (*5*). If so, a large fraction of the animals should have abnormal numbers of micronuclei. Our present data suggest that this is not so, as do also Kimball's (*1*) qualitative observations on the presence of macronuclear anlagen in the progeny of paramecia given doses of a few kiloroentgens.

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A Skeletal Difference Between Sublines of the C3H Strain of Mice

E. L. Green

Department of Zoology,
The Ohio State University, Columbus

The C3H strain of mice has been used in numerous studies of genetics, cancer, and physiology for the past 25 years. It is one of the old famous strains, ranking with DBA (= dba) and C57BL (= C57blk). The strain was started in 1920 by a mating of a male from the Little strain of dilute browns (= DBA) with a female from the albino mice of H. J. Bagg (*1*). Three main sublines of C3H mice have been established. One of these is Strong's own strain at Yale University; one is Andervont's strain at the National Institutes of Health, which was separated from Strong's strain about 1930; and one is Bittner's strain at the University of Minnesota, separated from Strong's strain about 1933.

Recently it has been discovered that these sublines of C3H mice are not all alike, at least with respect to skeletal type. The purpose of this paper is to give the evidence for this claim and to call this observation to the attention of those who are using C3H mice on the supposition that the various sublines are interchangeable, or nearly so.

The number of thoracic and lumbar vertebrae may be represented by a notation such as 13/5 or 13/6. The last lumbar vertebra is sometimes modified so that on one side it resembles a sacral vertebra. These intergrades are called "asymmetrical."

The skeletal type of C3H mice was first observed in a sample of 200 descended from mice obtained from Strong in 1937. About 96 per cent of these were 13/5, 3 per cent were asymmetrical, and 1 per cent were 13/6 (*2*). A second sample of 767 was produced from mice obtained from Bittner at the Jackson Laboratory in 1939. This sample gave a distribution of skeletal types almost identical with the first. The two samples were combined into a single sample of 967 (*3*). In 1948–52, three samples of C3H mice were obtained, all of which trace to the Andervont subline of C3H (more recently called the Heston subline, C3H/He). These three samples were similar, having about 96 per cent 13/6, but all were markedly different from the previous samples. In 1950–52, fresh samples of 144 and 75 from Bittner's C3H and C3H fostered on C57BL were obtained. These were not exactly like any of the preceding sublines, but did rather closely resemble the C3H mice obtained from Bittner and from Strong. In May 1952, a preserved litter of 7 C3H mice was obtained from Wilson. These were descendants of C3H mice received from Strong in 1947. All 7 were 13/5. The data are summarized in Table 1. For comparison, data are included on the CBA strain, which Strong derived from the same cross that produced C3H. The sample consists of 153 CBA mice, descended from mice obtained from Carter,

TABLE 1

DISTRIBUTION OF SKELETAL TYPES IN SEVERAL SAMPLES OF C3H MICE AND IN ONE SAMPLE OF CBA MICE

Strain	Origin	Thoracic/lumbar vertebrae (%)				Number
		13/5	Asym	13/6	Other	
C3H	Strong in 1937	96	3	1	0	200
C3H	Bittner in 1939 (= Bittner's Z)	97	2	1	0.1	767
C3H/He	Law in 1948	2	2	96	0.6	177
C3H/HeJax	Jackson in 1950	0	3	97	0	30
C3H/HeRl	Russell in 1950	1	3	96	0	72
C3H/Bi	Bittner in 1950 (= Bittner's Z)	93	6	1	0	144
C3Hb/Bi	Bittner in 1950 (= Bittner's Zb)	88	1	11	0	75
C3H/Wi	Wilson in 1952	100	0	0	0	7
CBA/Ca-*se*	Carter in 1950	95*	3	2	0	153

* Includes 10 *sese* mice of type 12/6 and 40 *sese* mice of type 13/5.