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DIFFERENTIAL LIFE SHORTENING INDUCED BY IRRADIATION WITH ELECTRONS IN SPECIES OF *DROSOPHILA*

RODOLFO FÉLIX E. and
JAVIER RAMÍREZ F.*

RESUMEN

Se describe el diseño experimental y el tratamiento matemático para la interpretación de los resultados, de la reducción de la longevidad inducida por el tratamiento con electrones de 1 MeV en especies del género *Drosophila* (*melanogaster, simulans, virilis* y *pseudoobscura*).

La curva de la viabilidad media de los controles y de los grupos tratados con dosis bajas sigue una distribución normal, mientras que los tratamientos con dosis altas dan curvas de distribución logarítmica normal. Al graficar la viabilidad media en función de la dosis también se obtiene una distribución normal para *D. melanogaster, D. simulans* y *D. virilis*.

SUMMARY

Several species of the genus *Drosophila* (*melanogaster, simulans, virilis* and *pseudoobscura*) were irradiated with 1 MeV electrons in order to analyze the reduction of the mean life span of each species. The experimental design and its mathematical treatment are outlined.

The mean life span of the controls as well as that from the groups treated with low doses follows a normal distribution, whereas a logarithmic normal distribution was found in the groups where high doses were applied. The relationship between mean survival time and dose also follows a normal distribution for *D. melanogaster, D. simulans* and *D. virilis*.

INTRODUCTION

Drosophila is an especially adequate organism for the research on the life shortening induced by irradiation, namely since the adult has little cell multiplication and consequently endures doses many times as high as the effective dose to kill larvae. As was stated by Müller (1960) the mortality in the adult *Drosophila* represents basically the same phenomenon as that referred to in the case of vertebrates as "aging by irradiation". In vertebrates the steadier course of the life cycle and of mitotic divisions makes the mortality more evenly spread out.

It is not yet well known to what extent the causes underlying the life shortening induced by irradiation in insects and mammals are the same, but the similarity of experiments and results in both groups justify the hopes that research on *Drosophila* may throw some light on the processes concerning ageing and radiation induced life shortening in mammals.

Lamb (1966) has suggested that the radiation injury causing life shortening in *Drosophila* is in part reparable and in part irreparable as it has been assumed for the acute and delayed effects of radiation on mammals. The absence of mitosis in the so-

* Programa de Genética y Radiobiología. Comisión Nacional de Energía Nuclear.

matic cells of the adult insect, makes it very resistent to irradiation in comparison to other animals as vertebrates, and it is a well known fact that the doses of radiation required to kill an adult insect are from a hundred to a thousand greater than those that will have lethal effects upon a vertebrate (Rockstein, 1966).

In *Drosophila melanogaster* (Berlin wild stock) the mortality rate after irradiation fits a normal distribution, which is altered at doses higher than 90 Kr (Nöthel, 1963). This change in the survival rate curve, as Nöthel indicates, seems to be due to a new type of mortality, which in agreement with the radiation syndrome is interpreted as a central-nervous-death. Therefore, when the mean lethal doses (LD_{50}) from several species are recorded on different days after the treatment data are obtained which may not be comparable.

Sonnenblick (1964) found a positive relationship between longevity and radioresistance in strains of *Drosophila melanogaster.* In view of the differences that may appear in the survival curves on several periods after irradiation due to the particular life span of each species, it is advisable to design a method that will allow a comparison on the survival rate after irradiation between the species which may differ in its characteristic longevity.

This paper will describe a method developed in order to obtain comparable data on the survival rate after irradiation of imagoes of several species of *Drosophila.*

MATERIAL AND METHODS

The species of *Drosophila* used throughout the experiment were reared in mass cultures in half pint bottles with the agar-cornmeal medium regularly employed in the laboratory. All the cultures were kept at a temperature of 25± 1°C before and after the treatment. Groups of 300 male and female flies from each species were irradiated as adults, 0 to 24 hours after emergence from the pupal stage. The imagoes were etherized and introduced in polyethylene containers for the electron treatment, and counting was done everyday until the last fly's death was recorded. It was necessary to change the cultures every five days in order to avoid the spoiling of the medium. No larvae were ever noted in cultures with irradiated adults.

The irradiation treatment was carried out at the Van-de-Graaff accelerator at the National University of Mexico. A flux of accelerated electrons of 8.32±0.70 K rad/sec with a current of 5 μA was applied at a distance of 25.00 cm from the window.

Four species of *Drosophila* were tested: *D. melanogaster, D. simulans, D. virilis* and *D. pseudoobscura.* The mean life span for each group and its control was determined in the following manner: a sigmoid graph (not included in the text) was obtained plotting accumulated mortality percentage against survival time. Using the probit transformation a second graph was drawn to situate on the time scale the point (mean survival time) corresponding to the mid point in the scale of mortality (Tab. I). The reason was calculated by dividing the mean survival time of each group by its control. The substraction of this value from the unit was done to obtain the reduction of the mean life span due to the treament. Graphs (Fig. 1) were drawn plotting the reduction of the mean life span against the dose, such sigmoid curves were transformed into straight lines applying the probit transformation; the LD_{50} fits mid point on the scale of reduction of the mean life span (Figs. 2-5).

RESULTS

A normal sigmoid curve corresponding to a normal distribution of the mean survival life span was found for the controls of the four species under analysis, as well as for the following groups:

D. melanogaster: all doses from 8.32 to 74.88 Krad
D. simulans: 8.32, 16.64 and 33.28 to .. 66.56 Krad
D. virilis all doses from 8.32 to 58.24 Krad
D. pseudoobscura: 8.32 and 58.24 Krad

In the following groups a sigmoid logarithmic curve was found corresponding to a logarithmic normal distribution of the mean survival life span:

D. Melanogaster: 74.88 Krad
D. simulans: 24.96, 74.88, 83.20 and 91.52 Krad
D. virilis: 66.56 and 74.88 Krad
D. pseudoobscura: 24.96 to 49.92 and 66.56 Krad

The distribution of the reduction of the mean life span againts the dose shows a normal sigmoid curve for the species: *melanogaster, simulans,* and *virilis.* For. D. *pseudoobscura* a logarithmic sigmoid was found for the same relation.

The analysis by means of the t-student procedure of the LD_{∞} values determined in this experiment proves that all the diferences between the four species are significant to a level of 0.95 except for the combination *simulans-melanogaster* which are closely related species.

CONCLUSIONS

The mean life span of the controls as well as that from the groups treated with low doses follow a normal distribution, whereas a logarithmic normal distribution was found in the groups where high doses were applied.

The reduction of the life span plotted agains the dose corresponds to a normal distribution curve for the species: *melanogaster, simulans* and *virilis. D. pseudoobscura* shows a logarithmic normal distribution for this same relation.

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TABLA 1

Reduction of the mean life span of adults of *Drosophila* species after exposure to electron radiation.

1 sec- 8.32 Kr
2 sec-16.64
3 sec-24.96
4 sec-33.28 Kr
5 sec-41.60
6 sec-49.92
7 sec-58.24 Kr
8 sec-66.56
9 sec-74.88
10 sec-83.20 Kr
11 sec-91.52

D. simulans

Doses	Control	1 Sec.	2 Sec.	3 Sec.	4 Sec.	5 Sec.	6 Sec.	7 Sec.	8 Sec.	9 Sec.	10 Sec.	11 Sec.
Mean life span in days.	34.83	26.97	22.88	19.56	19.50	16.89	17.32	11.80	8.10	8.49	6.21	3.79

D. virilis

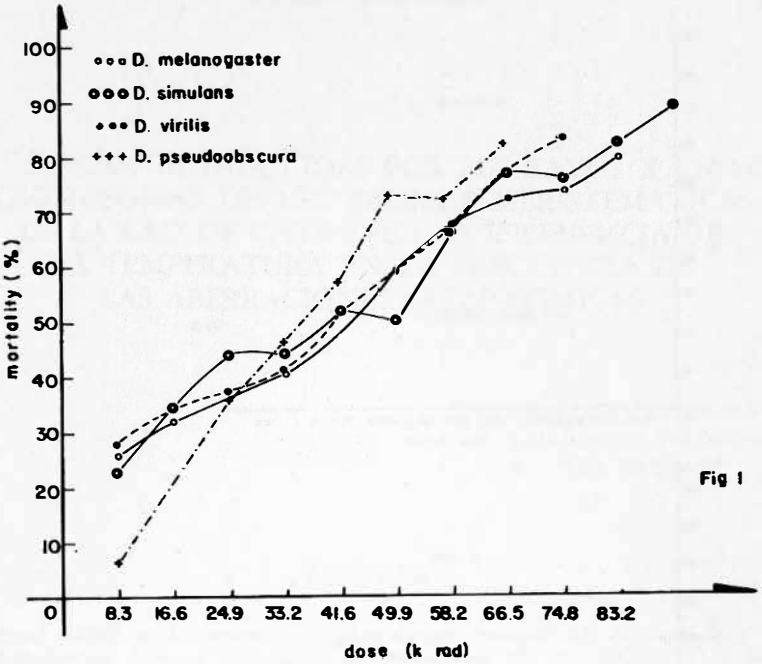
Doses	Control	1 Sec.	2 Sec.	3 Sec.	4 Sec.	5 Sec.	6 Sec.	7 Sec.	8 Sec.	9 Sec.	10 Sec.	11 Sec.
Mean life span in days.	54.03	38.79	35.50	33.71	31.68	26.16	16.64	17.87	12.29	9.06		

D. melanogaster

Doses	Control	1 Sec.	2 Sec.	3 Sec.	4 Sec.	5 Sec.	6 Sec.	7 Sec.	8 Sec.	9 Sec.	10 Sec.	11 Sec.
Mean life span in days.	36.47	27.00	24.68		21.59		14.83	11.82	10.09	9.62	7.35	

D. pseudoobscura

Doses	Control	1 Sec.	2 Sec.	3 Sec.	4 Sec.	5 Sec.	6 Sec.	7 Sec.	8 Sec.	9 Sec.	10 Sec.	11 Sec.
Mean life span in days.	32.31	30.29		20.80	17.40	14.19	9.05	9.51	5.26			



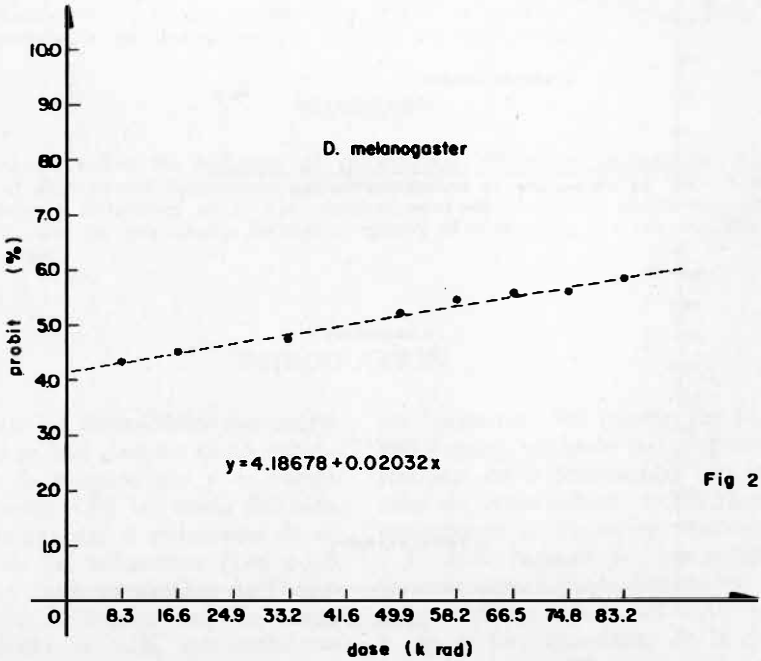
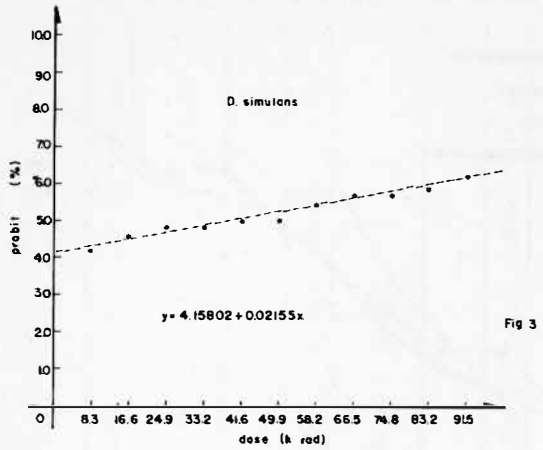
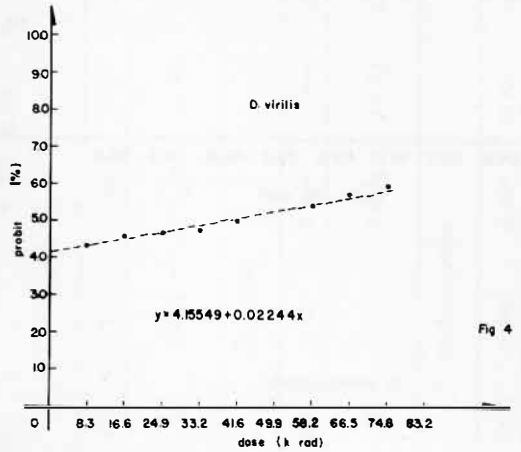


Fig. 1. Dose-response curves of *D. melanogaster, D. simulans, D. virilis* and *D. pseudoobscura*, for the reduction of the mean life span, expressed as per cent morality in the indicating ranges of exposure to fast electron irradiation.

Fig. 2. Probit transformation of the dose-response curve of *D. melanogaster* for the reduction of the mean life span in the range 8.32 to 83.20 K rad of exposure to fast electron irradiation.





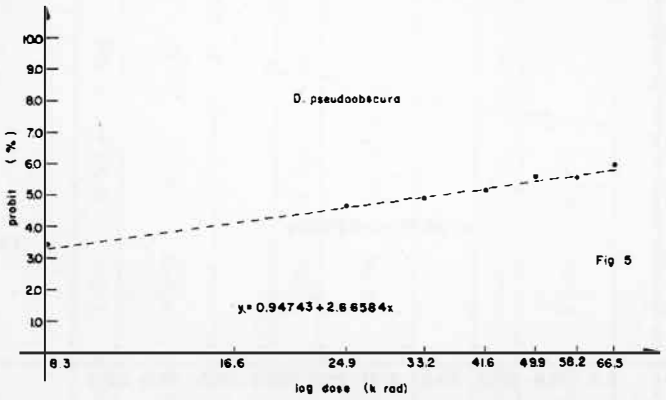


Fig. 3. Probit transformation of the dose-response curve of *D. simulans* for the reduction of the mean life span in the range 8.32 to91.52 K rad of exposure to fast electron irradiation.
Fig. 4. Probit transformation of the dose-response curve of *D. virilis* for the reduction of the mean life span in the range 8.32 to 74.88 K rad of exposure to fast electron irradiation.
Fig. 5. Probit transformation of the dose-response curve of *D. virilis* for the reduction of the mean life span in the range 8.32 to 66.56 K rad of exposure to fast electron irradiation.