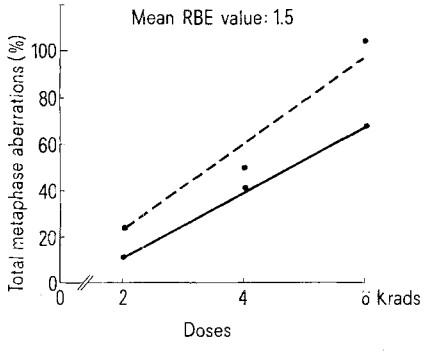
## Fast Rejoining Processes in *Nigella damascena* Chromosomes After Fractionated Exposures to Accelerated Electrons

In previous experiments with the plant species *Nigella damascena*, it was found that fractionation of $\gamma$-ray exposures results in a highly significant decrease of the frequency of lesions belonging to the chromosome class at time intervals longer than 2 min 10 sec[1]. This decrease was suppressed or reversed at exactly the same time interval when seeds were pretreated with a solution of various chelating agents. From these data, it was inferred that ionic bonds are involved in the fast rejoining processes[1].

These new experiments were designed to investigate if the conclusions drawn from experiments with $\gamma$-rays can be extended to accelerated electrons.




RBE's of accelerated electrons versus $^{60}$Co $\gamma$-rays (10 root meristems/20 metaphases per meristem). Dotted line: accelerated electrons. Full line: $^{60}$Co $\gamma$-rays.

Table I. Effects of fractionated exposures of accelerated electrons (2 × 2 krads) separated by increased time intervals

| Treatments | Time intervals (min) | | | | | |
|---|---|---|---|---|---|---|
| | 0 | 1 | 3 | 6 | 30 | 60 |
| Control | 32.5 | 20.0 | 17.5 | 22.5 | 16.5 | 21.5 |
| DP | 32.5 | 29.5 | 25.0 | 21.5 | 25.0 | 22.5 |
| DIECA | 20.5 | 23.0 | 18.5 | 18.0 | 28.0 | 23.5 |

Percentages of chromosome aberrations (10 root meristems/20 metaphases per meristem).

Table II. Effects of fractionated exposures of accelerated electrons (2 × 2 krads) separated by increased 10 sec intervals

| Treatments | Time intervals (sec) | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | 0 | 10 | 20 | 30 | 40 | 50 | 60 | 70 | 120 |
| Control | 48.5 | 53.5 | 48.5 | 33.0 | 32.0 | 33.5 | 32.5 | 31.0 | 33.5 |
| DP | 28.5 | 32.0 | 39.0 | 31.0 | 31.5 | 30.0 | 27.0 | 23.5 | 33.0 |
| DIECA | 31.0 | 36.0 | 31.0 | 42.0 | 45.0 | 39.5 | 42.0 | 43.5 | 42.5 |

Percentages of chromosome aberrations (10 root meristems/20 metaphases per meristem).

*Material and methods.* The material *Nigella damascena* dry seeds has been previously described in detail[2]. Seeds (about 8% water content) were irradiated in a Betratron with 34 MeV electrons at 3 dose rates in 3 series of experiments, respectively 740 rads/min, 674 rads/min and 847 rads/min. The seeds were irradiated by the electron beam in the center of a plexiglas block 3 cm thick backing the monitoring chamber of the electron beam of the Betraton, Brown-Boveri 35 Asclepitron.

The absorbed dose relative to water was derived from ionization measurements with a 0.6 cm$^3$ Baldwin chamber in the irradiation site of the seeds and checked by absorbed dose measurements with a Fricke dosimeter. A supplementary verification was made by the same methods in the same sample holder at the end of the diaphragm defining a field of 12 × 12 cm. The dose rates mentioned are the effective ones obtained for the different irradiations and the precision ranges vary from 3.6% to 5.1% at the 95% confidence level.

The first series, designed to evaluate the relative biological effectiveness (RBE) of accelerated electrons, was compared with an irradiation with $^{60}$Co $\gamma$-rays (dose rate 362 rads/min at 25 °C, 53% relative humidity) at doses ranging from 2 to 6 krads. In the $\gamma$-ray beam, the seeds were irradiated in the center of the same plexiglas block set close to the collimator, at full aperture, of an about $^{60}$Co 2500 Ci Picker C8M/80 installation and backed by a 10 cm thick plexiglas block.

The second series was designed to investigate the effects of fractionated exposures of accelerated electrons. The 2 exposures (2 × 2 krads) were separated by increased time intervals ranging from 1 to 60 min.

The third series of experiments was made to confirm and to precise the results of the second. The 2 exposures were separated by time intervals ranging from 10 to 120 sec with a regular increment of 10 sec (except for the last interval). Seeds were sown immediately after irradiation and the chromosome damage observed during the first mitotic cycle after irradiation. The cytological procedures have been reported elsewhere[3].

For experiments with chelating agents, seeds were treated either by sodium diethyldithiocarbamate (DIECA) or dipyridyl (DP) ($1 \times 10^{-4}$ $M$/8 h at 20 °C) then redried to the initial moisture before irradiation.

*Results.* The Figure summarizes the results of the first experiments. RBE's of accelerated electrons versus $^{60}$CO $\gamma$-rays are of the same order of magnitude as those reported by other researchers for quite different materials. These data allowed us to select the appropriate exposures for the experiments of fractionation.

Table I shows a significant decrease of the number of aberrations between 0 and 1 min interval i.e. non fractionated and fractionated exposures (the F value between 0 and 1 min is 12.18**). The threshold value (−40%) is comparable to the mean threshold value obtained with $\gamma$-rays (−41.3%)[1]. However, the data suggest that the drop occurs earlier after accelerated electrons than after $\gamma$-rays. As for $\gamma$-rays, the drop is suppressed by both chelating agents (statistical differences not significant).

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Table III. Effects of fractionated exposures of accelerated electrons on different types of aberrations (2×2 krads)

| | Time intervals (sec) | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | 0 | 10 | 20 | 30 | 40 | 50 | 60 | 70 | 120 |
| Breaks | 10.5 | 15.5 | 14.0 | 11.5 | 8.5 | 11.0 | 7.5 | 8.5 | 7.5 |
| Asymmetrical exchanges | | | | | | | | | |
| Interchanges | 16.5 | 18.0 | 17.0 | 10.5 | 11.0 | 13.5 | 11.5 | 10.5 | 16.5 |
| Intrachanges | 7.5 | 7.5 | 7.0 | 5.5 | 2.5 | 3.5 | 4.0 | 2.0 | 4.5 |
| Minutes | 14.0 | 12.5 | 10.5 | 5.5 | 10.0 | 5.5 | 9.5 | 10.0 | 4.5 |

Percentages of aberrations (10 root meristems/20 metaphases per meristem).

The results of the third series are given in the Table II. This shows that decrease occurs between 20 and 30 sec time interval (F 20–30 sec = 21.7**) and no decrease appears after chelating agent treatments (contrariwise, a slight increase for DIECA is seen between the same time intervals).

Table III shows that all kinds of aberrations: breaks and exchanges are involved in the decrease. Minutes (fragments smaller than 0.5 $\mu$) were classified separately, owing to the uncertainty of their origin. Symmetrical exchanges were not taken into consideration on account of the difficulty of detection of such lesions. In the class of dicentric chromosomes, the proportion of dicentrics accompanied with 2 acentric fragments can be generally considered as an index of incomplete rejoining. This proportion was significantly much lower after the critical interval. A score of 100 dicentrics yielded the proportion 20% before, against 4% after ($\chi^2 = 10.7$, $p \simeq 0.001$).

*Discussion and conclusion.* The time interval after $\gamma$-rays (2 min 10 sec) is the minimum delay necessary for the suppression of interaction between both exposures due to fast rejoining processes[1]. In the present experiments, we proved that these processes also operate after accelerated electrons. The fact that the minimum delay before the drop is shorter than after $\gamma$-rays (20 sec instead of 2 min 10 sec) could be due to differences of experimental conditions, especially the higher dose rate in the case of accelerated electrons.

The two chelating agents react in the same manner as after $\gamma$-irradiations i.e. by suppressing the decrease obtained in the series without chelating agents. The decrease of the damage is generally thought to be due to the preponderance of restitutions. Some of those 'restituted breaks' could not participate in exchanges, which explains the decrease of the number of aberrations. In the present experiments, this statement was confirmed since the proportion of incomplete exchanges was considerably decreased after the critical time interval. This relative decrease of incompleteness would be a biased estimate of the chromosome break restitution.

*Résumé.* Des graines «sèches» de nigelles ont été irradiées par des doses fractionnées (2×2 krads) d'électrons accélérés (34 MeV). Une diminution très significative des taux de toutes les lésions chromosomiques a lieu entre les intervalles de temps 20 et 30 sec. Après un traitement des graines par 2 agents de chélation (DP et DIECA), il y a suppression de l'effet.

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## Evolution in a Cosmopolitan Species: Genetic Latitudinal Clines in *Drosophila melanogaster* Wild Populations

*Drosophila melanogaster*, a widespread cosmopolitan species, if often considered of low interest for evolutionary studies. This is mainly due to the fact that these flies are ecologically linked to human agricultural activities[1,2]. It is generally assumed that *melanogaster* was introduced by man in many countries and that genetic exchanges between foreign populations are still favored by fruit transports. Thus, recent introduction in many countries and permanent mixing of populations should leave little time for the appearance of geographic divergences and the formation of local races. We leave open the question of the extent of man's influence on *melanogaster* genetics, but show that strong latitudinal selection exists among these flies.

TEISSIER et al.[3–8] showed that quantitative biometrical differences distinghish French and Japanese strains. This conclusion resulted from the study of numerous strains, but the frequency distributions of different strains of the same geographic origin overlapped greatly so that it was not possible to determine their individual origins. The reason for this now appears to be a genetic drift which artificially increased variability between strains kept under laboratory conditions for several years[9,10]. For most quantitative biometrical characters, freshly caught flies show less heterogeneity.

More recently, strains from tropical Africa reared in standard 25 °C laboratory conditions were found to be much smaller and to have fewer ovarioles than French

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