Technical Papers

Time-Intensity Factor in the Production of Dicentric Bridges with Gamma Rays of Radium during Meiosis in the Grasshopper, *Gesonula punctifrons*

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A large number of dicentric bridges are observed in the first anaphase of meiosis, when the resting cells of the primary spermatocytes of the grasshopper, *Gesonula punctifrons* (*1–3*), are treated with moderate doses of x-rays. These bridges (Fig. 1) are morphologically indistinguishable from those produced as a result of crossing over within relatively inverted segments of chromosomes. Ray-Chaudhuri and Sarkar (*3*) have, however, found the frequency of dicentric bridges in meiosis to be directly proportional to the dose of x-rays, tested within the range of 40 to 320 r units. In view of these findings, they concluded that the experimentally produced bridges in *Gesonula* are not the result of crossing over within an inversion, since the production of an inversion requires at least two independent breaks, but that, instead, they originated from a single break in a meiotic chromosome or, less probably, from nearby breaks produced in two chromatids by the track of one and the same ionizing particle.

A reliable test for the single unit type of action of radiation is to find whether the effect of a given dose is independent of the manner in which the ionizations are distributed in time. The present series of experiments was designed to find the effect of varying the intensity (dose-rate) of treatment on the frequency of dicentric bridges in the same material (*4*).

In this, as well as in the previous study conducted by Ray-Chaudhuri and Sarkar (*3*), adult males of *Gesonula punctifrons* were used as the experimental material. In each of the five experiments carried out, the grasshoppers were divided into two equal lots; one lot was irradiated for 30 min and was kept during irradiation in a rectangular cardboard box 4 by 2 by 1 cm in size, and two radium needles of 50 mg each were kept at a distance of 2 cm from the middle of the box. The other lot was treated for 23 hr and was kept in an annular box 1 cm wide and 2 cm high, with a mean radius of 10 cm. Two radium needles of 21 mg each were placed at the center of the box. Under these conditions of radiation, the calculated dosage for both lots of grasshoppers was 81.6 r units. Thus, a difference in intensity amounting to 46 times was produced between the high- and low-intensity lots. The testes were fixed 30 hr after the completion of irradiation in each case, and results were scored from temporary acetocarmine preparations.

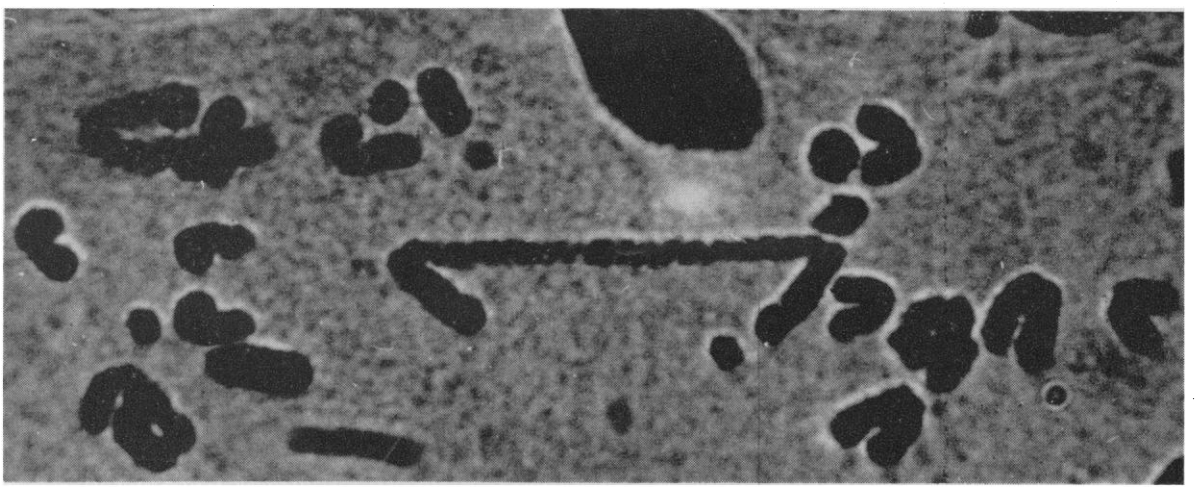

Fig. 1. Photomicrograph of a typical dicentric bridge with fragment; 80 r (high intensity).

In all five experiments, a total of 9679 first anaphase cells were examined and 408 bridges were recorded, from 90 treated grasshoppers. The results are summarized in Table 1. It will be clear from the table that the frequencies of bridges are remarkably similar in all five experiments, both in the high- and low-intensity lots. The differences in the percentage of bridges produced after high- and low-intensity irradiations, either in the individual experiments or in the total of all experiments, are insignificant. We, therefore, conclude that this particular type of radiation-induced effect in *Gesonula* is independent of the intensity of radiation within the limits of the experiments.

The results of the present series of experiments, coupled with the fact that the frequencies of bridges are in direct proportion to the x-ray doses, further

Table 1. Frequency of dicentric bridges 30 hr after irradiation in different high- and low-intensity experiments with 80 r of gamma rays.

Expt	High intensity			Low intensity		
	First anaphase cells	Bridges	Percentage	First anaphase cells	Bridges	Percentage
I	384	15	3.90	830	34	4.09
II	1175	58	4.94	474	23	4.85
III	492	23	4.67	502	20	3.98
IV	2025	85	4.20	2073	80	3.85
V	919	40	4.35	805	30	3.72
Total	4995	221		4684	187	
Mean %			4.42			3.99

strengthened the conclusion that the bridges originated not through two independent breaks in a chromosome but largely as a result of a single break in an unsplit chromosome, caused by a single ionization track.

The sensitivity of the chromosomes of the primary spermatocyte resting cells of *Gesonula* to x-ray breakages does not alter appreciably for a relatively long period, inasmuch as Ray-Chaudhuri and Sarkar (*3*) have demonstrated the constancy of the frequency of first anaphase bridges recorded at different hours after irradiation up to a period of 148 hr. It is also likely that there is a similar stable period in the meiotic resting cells of plants. It has recently been shown by Darlington and LaCour (*5*) that in *Tradescantia bracteata* a constant frequency of first anaphase bridges is found between 16 and 48 hr after irradiation with x-rays. They, however, interpreted these bridges as originating from sister union of broken ends of chromatids (physiological effect) and not from chromosome breakage.

But the dicentric bridges in the meiotic cells of *Gesonula* are undoubtedly due to breakage in the chromosomes and not to any kind of physiological disturbance. This can be substantiated by the following evidence. An examination of the numerous bridges obtained by us during the course of our experiments reveals that the length of the dicentric portion of the chromatid between the two centromeres varies in different nuclei, in such wise that the length of the accompanying fragment (Fig. 2*a*, *m*) has a negative correlation with the length of the dicentric portion of the bridge (Fig. 2*a*, *l*). From the size of the monocentric free arms at the two ends of the bridge (Fig. 2*a*, *n*), we can get an idea about the size of the bivalent. A comparatively short dicentric portion relative to the size of the unaffected chromatid (free arm) indicates a break near the centromere. A bridge resulting from such a proximal break will be accompanied by a comparatively large fragment (Fig. 2*a–d*). On the other hand, the bridge originating from a distal break (Fig. 2*e–h*) will have a large dicentric portion and a small fragment. These conditions are always satisfied in all the bridges examined by us from this point of view.

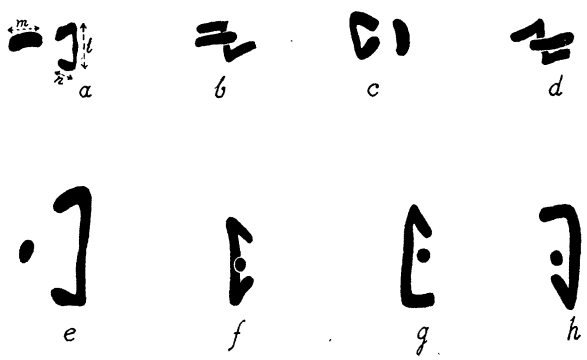

Fig. 2. *a–d*, dicentric bridges resulting from proximal breaks; *e–h*, bridges resulting from distal breaks.

References and Notes

1. B. P. Uvarov of the British Museum (Natural History) has recently informed us that the correct name for this species should be *Gesonula* (and not *Gesonia*) *punctifrons* as used by Ray-Chaudhuri and Manna (*2*) and Ray-Chaudhuri and Sarkar (*3*).
2. S. P. Ray-Chaudhuri and G. K. Manna, *J. Expt. Zool.* **114**, 421 (1950).
3. ——— and I. Sarkar, *Science*, **116**, 479 (1952).

4. We are deeply grateful to H. J. Muller, of Indiana University, for valuable suggestions incorporated in this paper. Thanks are also extended to S. Mitra, Director, Chittaranjan Cancer Hospital, for providing facilities for irradiation in his hospital and to A. Bose, physicist of the same hospital, for his help in arranging the irradiations.

5. C. D. Darlington and L. F. LaCour, *Heredity Suppl.* **6**, 41 (1952).

Received February 19, 1954.

Quantitative Flocculation of *S. schottmuelleri* Cells by Quaternary Ammonium Germicides[1]

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In recent years, relationships have been shown to exist between bactericidal properties and certain physico-chemical properties of quaternary ammonium germicides, for example, the release of conducting material by quaternary-treated cells (*1*) and the adsorption of quaternaries on wool (*2*). This report describes an attempt to relate bactericidal activities of quaternaries to the property of these agents of causing macroscopic flocculation of the test organisms. Clumping of bacteria as a result of the action of the quaternaries has been observed microscopically (*3*), but we are aware of no previous attempts to develop this observation into a macroscopic method for studying antibacterial action.

The addition of graded levels of quaternary germi-

[1] The opinions expressed herein are those of the authors and are not necessarily similar to the views of the Department of the Navy.