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RADIATION-INDUCED CHROMOSOME ABERRATIONS AND LOSS OF REPRODUCTIVE INTEGRITY IN *TRADESCANTIA*[1]

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Chromosome Aberrations and Survival

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INTRODUCTION

Much information has been accumulated in recent years on the loss of reproductive integrity in irradiated populations of animal cells. Data have been acquired mainly from studies of cell populations cultured *in vitro* and though such results can vary due to different culturing conditions (1,2) a comparison with the few results available from *in vivo* studies (3-5) has shown good agreement between the two sets of data. With higher plant cells, *in vitro* studies have not been technically feasible up to now and it has only been possible to estimate radiation induced loss of reproductive integrity either by studying the sequence of changes in the histological structure of an organized meristematic region (6) or by following the patterns of recovery of growth in irradiated roots (7,8). For most of these cell types--both animal and plant--the survival curves after x- and gamma-ray exposures are sigmoid, fitting the expression $S = [1 - (1-e^{-kD})^n]^m$, where S is the survival at dose D, *n* is assumed to be the number of sites of one particular type which must be inactivated to sterilize the cell (extrapolation number) and *m* is the number of different kinds of sites. *m* is generally assumed to be one, though this assumption has been disputed by Bender and Wolff (9) and Bender and Gooch (1). 1/k or D_o is the dose required to reduce the surviving fraction S to 0.37 S, i.e. ($e^{-1}S$) on the exponential part of the curve and has been found to be between 50 and 300 r for most of the mammalian cells investigated and between 35 and 90 rads for *Vicia faba* (7,8). Puck (10-12) on the basis of the good agreement of the mammalian cell survival curves to the equation

$$S = e^{-kD}(1 + kD) \qquad (1)$$

of the high values of k and also on the basis of cytological observations, has suggested that two- or multiple-hit chromosomal aberrations are one of the prime causes of reproductive death in mammalian cells. However, the validity of equation (1) for the mammalian cell survival curves obtained and of the relevance of certain of the cytological data have been questioned, and the conclusion of the prime importance of two-hit aberrations disputed by Bender and Wolff (9).

As has been repeatedly stated, the only definitive conclusion that can be drawn at present from numerous radiobiological experiments is that lesions induced within the nucleus are of prime importance in determining the loss of proliferative ability. Inhibition of mitosis, nucleolar inactivation, spindle inhibition and nuclear membrane damage are among some of the mechanisms which have been considered as reasonable alternatives to that of loss of genetic information. In the present paper, an experiment is described in which accurate assessments of chromosomal damage were made on a system in which data on reproductive integrity could also be obtained, and this has allowed a reexamination of the possible relationship of these two events.

EXPERIMENTAL MATERIAL

Clonal material (clone 02) of a diploid (2n = 12) variety derived from Tradescantia occidentalis[2] was utilized for the experiments. This clone is heterozygous for certain marker genes and is being extensively used in other experiments. The tissues involved were hairs produced within the flowers. Each inflorescence comprises a large number of serially

arranged buds of different developmental age. Under optimum conditions of growth, one flower emerges daily. Within each flower, there are six filaments, each bearing from 50 to 90 hairs. This hair number is constant for any given inflorescence but unless environmental conditions are kept uniform, varies from inflorescence to inflorescence. Plants were kept in a constant environment chamber (24 hours light, 950 foot candles, 70 ± 1° F) for seven days pre- and three days post-irradiation. Each hair is a chain of single cells and its pattern of growth is as follows: a cell in a filament divides in such a way that one of the daughter cells protrudes obliquely out of the filament. This protruding cell is meristematic and becomes the terminal cell of the hair, continuing to divide until the mature hair (20 to 30 cells) is formed. The subterminal cell always divides once after its formation and occasionally an intermediate cell also divides. Thus the hair is almost entirely the product of the single terminal cell.

DETERMINATION OF REPRODUCTIVE INTEGRITY

The rationale of the technique was that if flowers having immature hairs (from 2 to 7 cells long) were irradiated, then, since the full growth of the hair was dependent on the meristematic activity of the terminal cell, an inactivation of this cell should result in an aborted or stunted hair. The expected number of hairs per filament (the control value) was determined from the average value of three to four flowers (18 to 24 filaments) taken prior to irradiation from every inflorescence used. Percentage survival was then determined by scoring the number of normal hairs in ten to twenty flowers (that is, 60 to 120 filaments) on the 13th to 16th day post-irradiation.

CHROMOSOMAL ABERRATIONS

Terminal and subterminal hair cells were again used for determining aberration frequencies. In order to obtain a representative picture of the sensitivity of the asynchronous population of these meristematic cells, samples were fixed at the following intervals post-irradiation: 2, 4, 6, 8, 10, 13⅓, 16, 18½, 21, 24 and 26½ hours. Tissues were pretreated with 0.075% colchicine for two hours prior to fixation and 50 metaphase cells were examined in Feulgen stained squash preparations from coded slides at each of the fixation times. All chromatid and chromosome aberrations were scored.

RADIATION TREATMENTS

All treatments were given at room temperature with a G.E. Maxitron X-ray machine operated at 250 kvp and 6 ma. With the 1 mm aluminum filter used, the dose rate was 108 r per minute, the doses being measured with a Victoreen dosimeter placed on the turntable in the position occupied by the inflorescences. For the aberration frequency determinations, doses of 36, 63 and 90 r were used. At higher values the level of damage and the complexity of the configurations rendered interpretation difficult. For the assessment of reproductive integrity, doses of 85 to 500 r were used. Above 500 r, the flowers were so badly injured that they did not develop and thus the level of damage could not be assessed.

RESULTS

Chromosome and Chromatid Aberrations

The frequency of the different forms of chromatid and chromosome aberrations, as well as that of achromatic lesions (13), was determined at all fixation times after the three doses of radiation and are given in Table I. At the two lowest doses used, there seemed to be little fluctuation in sensitivity over the mitotic cycle in terms of the production of chromatid aberrations, even up to 26½ hours post-irradiation. The normal mitotic cycle time is around 20 hours (Steffensen, personal communication), a value similar to that obtained in *Tradescantia* roots (14) where the average duration of the pre-synthetic (G_1), DNA synthetic (S), post-synthetic (G_2) and mitotic (M) periods have been shown to be 4.0, 10.8, 2.7 and 2.5 hours respectively. Chromatid aberrations are usually produced only after irradiating G_2 and S cells. The appearance of chromatid aberrations even 26½ hours post-irradiation must therefore mean that the radiation induced a considerable retardation in the movement of G_2 and possibly S cells through the mitotic cycle.

Chromosome aberrations were observed in relatively low numbers and so the data relating to their frequency cannot be considered too reliable. In general, chromosome aberrations are much less frequent and their importance is probably much less than chromatid aberrations since the G_1 period occupies a small fraction of the cycle--estimates vary from 4 hours (14) to possibly as little as one hour in *Tradescantia* roots (15). It might have been considered desirable to make fixations at longer times after irradiation in order to obtain a better assessment of the frequency of chromosome

aberrations, but then the problem of the appearance of mixed populations of X_1 and X_2 cells at mitosis arises. It is important that only X_1 cells be examined since badly damaged cells tend to be eliminated in later generations. The only cell which could be identified with certainty as being an X_2 cell appeared at 26½ hours after 63 r. With the 90 r treatment a definite peak of chromatid aberrations, especially interchanges, occurred at the early fixation times (Table I). Since these interchanges have a large "dose squared" component (Table II), any fluctuations in sensitivity throughout the cell cycle would be much more apparent at the highest dose. A peak aberration frequency in late interphase cells of Vicia faba has been observed by Evans (personal communication) and presumably the 4 to 8 hour peak in the present observations represent the G_2 population of cells, if mitotic delay is taken into account.

The question of the possible confusion of chromatid breaks and gaps has been considered at length by Revell (13,16) and an attempt was made to separate these two types of events in the present experiment. The frequency of gaps obtained was very high (see Table I) and compares well with the values reported by Revell but the frequency of chromatid breaks was also somewhat high. Revell (13) has suggested that all chromatid aberrations may be the result of exchange events and on the basis of certain assumptions has predicted that the ratio of chromatid breaks to incomplete isochromatid breaks should be 2.5:1. He observed ratios of 3.2:1 and 2.5:1 in two separate experiments with Vicia faba roots (16), and Evans (17) has similarly reported ratios of 5.2:1 and 3.5:1 for Vicia faba roots and Tradescantia pollen tubes respectively, all the values being in agreement with that

expected. In the present data the overall ratio was 3.3:1 (256 chromatid to 84 incomplete isochromatid breaks). The level of incompleteness was somewhat higher (27.1% for isochromatid aberrations and 19.7% for exchanges) than the values of 5-10% obtained by Revell (13,16) but not very different from those obtained by Conger (18) (18%) and by Neary and Evans (19) (30%). Nevertheless, in spite of the apparent agreement in the expected ratio of chromatid to incomplete isochromatid breaks, the frequencies of chromatid breaks may possibly have been overestimated. Certainly the level is considerably higher than that quoted by Revell (16) and by Bender (20) though not very different from those quoted by Neary and Evans (19) and Chu _et al._ (21), all of whom similarly attempted to exclude gaps from their data. This reservation regarding the chromatid break data will be considered in a later section.

The equations which best fit the lines relating aberration frequency to dose are given in Table II. The isochromatid data best fit a linear equation but the chromatid break and interchange data better fit a quadratic than a linear equation. Again, on the exchange hypothesis of aberration induction, at sufficiently high doses all aberrations might be expected to have a "dose squared" component. It must be remembered that the accuracy of this dose response relationship may not be very great as different cell populations are probably represented in the period 0-26½ hours after the three different doses, due to the varying amounts of mitotic delay induced. Nevertheless, it is important that a representative picture of the sensitivity of the whole spectrum of stages of the mitotic cycle be obtained if any attempt is to be made to equate aberrations to loss of

reproductive integrity. Poisson tests of the distribution of all chromatid aberrations per cell, and also of chromatid exchanges per cell, showed good agreements with expectation, indicating that the more badly damaged cells were not being selected against in their movement into metaphase.

Reproductive Integrity

The percentage of hairs which continued to grow to a normal length after irradiation, that is, the number of irradiated terminal cells which could divide at least 5 or 6 times, are given in Fig. 1. This criterion of ability to divide at least this number of times, is the usual one adopted in scoring viability by colony counts in *in vitro* experiments with mammalian cells. Reproductively sterile apical cells either did not divide or divided only once or twice post-irradiation. As in other irradiated cell populations, at the higher dose levels more cells tended not to divide at all after irradiation. Giant cells with large nuclei arose fairly commonly. The extrapolation number determined from a regression analysis of the points on the exponential part of the semi-log plot (Fig. 1) was 1.6, the D_o value 148.7 r.

DISCUSSION

The parameters defining the reproductive integrity of the irradiated apical cells, $n = 1.6$, $D_o = 148.7$ r, were remarkably similar to those obtained from a diversity of mammalian cells, but somewhat different from those reported for the only other higher plant investigated, *Vicia faba*. For this latter species, values of $n = 2\text{-}3.5$ and $D_o = 35\text{-}90$ rads were obtained (7,8). In terms of gamma radiation-induced reduction in growth,

Vicia faba has elsewhere (22) been shown to be more resistant than *Tradescantia*. In making any such comparisons, it has to be borne in mind that there is one essential difference between the mechanics of the system involved in the present experiment and those involved in *in vitro* experiments. The criterion of integrity used here was the ability of one specific terminal cell to continue dividing. In *in vitro* experiments, and possibly in many other, though not all tissues *in vivo*, the criterion of integrity is met if either one of two daughter cells continues to divide. Though in many tissues, on average for every cell division that occurrs, one stem cell and one differentiated cell is produced, the tissue organization is not such that after every division one daughter cell remains meristematic and the other differentiates. This difference between the terminal cell situation and other cell populations is best illustrated by considering the fate of an induced lethal event in a particular cell. In the terminal hair cell, there is a 50% chance that this lethal event will segregate to the terminal cell, so that hair growth cannot continue, whereas with colony development *in vitro*, where all cells are potentially proliferative, a viable colony will be scored if any one of the daughter cells of the treated cell survives. Thus, if lethal events are induced which can segregate, the estimate of lethality will be higher with the present system than with *in vitro* techniques.

In attempting to equate loss of genetic information to lethality, the first and simplest assumption that can be made in any calculation is that any loss of chromatin will result in lethality. On this basis the fate of each cell examined for aberrations in the present experiment was determined,

assuming that there was a 50% chance of the apical cell escaping a deficiency after a chromatid break, no chance after an isochromatid break, 25% chance after an asymmetrical chromatid interchange, etc. (23). Minutes were ignored. From the data, 36 r would then be expected to give 79% survival, 63 r 68%, and 90 r 53.3%. Since there was some degree of uncertainty associated with the scoring of chromatid breaks (see above), the same calculation was again made ignoring these aberrations and the new values were 83%, 71% and 60% respectively. These two sets of values do not resemble the observed ones (Fig. 1) indicating that many deficient cells are reproductively viable.

In another attempt to equate the chromatid aberration and survival data, the equations relating yield to dose, $Y = \alpha \text{ iso } D$, $Y = \alpha \text{ chr } D + \beta \text{ chr } D^2$ and $Y = \alpha \text{ int } D + \beta \text{ int } D^2$ (Table II) where iso = isochromatid break, chr = chromatid break and int = chromatid interchange, were used to calculate the expected mean yields of total aberrations per cell at the doses used in the survival experiment. The other aberrations which were quantitatively less important were ignored. Knowing these mean values and assuming a Poisson distribution, the proportion of cells with different numbers of aberrations were calculated from the equation

$$\frac{\mu^x e^{-\mu}}{x!},$$

where μ is the mean value per cell, and x is the number 0, 1, 2, 3...... of aberrations per cell. (It was known that the distribution of total aberrations per cell at 36, 63 and 90 r were distributed in a Poisson manner.) If the cells with no aberrations or even cells with one or less aberrations were assumed to be reproductively viable (both including and excluding chromatid

breaks), then the predicted "survival" values were very much less than the observed (Table III).

Another assumption that could be made was that loss of reproductive integrity occurred when any two homologous chromosome arms among the 12 chromosomes of the diploid complement were simultaneously damaged. The frequency with which such damage would occur (assuming completely random distribution of aberrations between chromosome arms) at any given mean aberration frequency per cell was calculated, but again the predicted "survival" was much higher than the observed (Tables IVa and IVb).

There may well be a basic false assumption involved in these last two calculations (Tables III and IV)--that the yield of aberrations per cell increases with dose as the equation $Y = \alpha D$ or $Y = \alpha D + \beta D^2$ would predict. The predicted values would not be achieved if there was a limitation of the number of aberrations that can be produced in any given cell. The concept of a limited number of sites within the cell at which exchanges can occur has been considered by Wolff (24). A test for such a limitation of site number, using the chromatid exchange data at 90 r, showed that a Poisson distribution fitted better than a binomial distribution $((1 - \frac{m}{n}) + \frac{m}{n})^n$ at least with values of n (site number) up to 4. It is still possible, however, that there is a low number of sites and thus a limit to the number of aberrations per cell which can occur at high doses.

The percentages of cells free of aberrations at 36, 63 and 90 r were 71, 57 and 41 respectively--in other words the values appeared to decrease in an exponential manner with increasing dose within this range. Thus a further approach to the present problem was to predict the chromosome

damage which would occur at high doses from the mean aberration dose, i.e. that dose that will give on average, one scorable aberration per cell (9, 10). This value was obtained using the three equations in Table II--again aberrations other than these three main types were ignored. Y is given a value of 1 and D is calculated. This mean aberration dose, 96.2 r was then taken to be the 37% dose and the Poisson formula with $\mu = \frac{D}{D_o}$, where D_o is the 37% dose, used to calculate the expected frequency of cells with 0, 1, 2, 3...... aberrations per cell at different levels of D. If the proportion of cells assumed from this calculation to have one or less aberration were equated to survivors, then there was a remarkably good fit of predicted to observed survival (Table V). This calculation, however, involved the unlikely assumption that the number of cells free of aberrations did decrease in an exponential manner with increasing dose. If there is a limitation on aberrations per cell due to site number of other limitations, this expected relation may hold, but if there is no limitation, the exponential relationship as discussed above will not be found since the "dose squared" component becomes progressively more important at higher doses.

One further possible link between aberration and lethality may be that of bridge frequency. Bridges may cause mechanical difficulty at mitosis and often lead to genetic loss either directly or through the setting up of breakage-fusion-bridge cycles. Frequencies of cells which might be expected from the configurations present at metaphase to have one bridge and of those with two or more bridges are given in Table VI. The frequencies of the latter, but not the former, did bear some resemblance to the survival values, but it was impossible to extrapolate from the frequencies given in Table VI

to higher doses as there was no indication of the shape of the dose response curve. This posed the same problem as had been encountered previously. At those doses where survival could be accurately estimated, it was difficult if not impossible to obtain accurate detailed observations of aberrations.

A comparison of the present results with the chromosome aberration data of Bender and Wolff (9) and Chu _et al_. (21) is interesting. For diploid (2n = 46) human epitheloid cells, Bender and Wolff quote a mean aberration dose of 220 r. For diploid human, primarily fibroblast cells, the data of Chu _et al_. give a value of approximately 78 r and for _Tradescantia_ (2n = 12) the value is 96 r or if chromatid breaks are neglected, 129 r. At these mean break doses, the level of survival is generally lower for the diverse types of human cells which have been examined (some of them having a higher chromosome number than 46), though the relative amount of genetic information lost per break will be in the ratio of 1/46 or less:1/12 for the human and _Tradescantia_ lines respectively. This would argue against the primary importance of single aberrations at least in determining lethality. That some considerable degree of genetic loss can be tolerated even in a diploid cell is well known and has been demonstrated again in the present study. That loss of genetic information contributes to reproductive sterility in many instances is also obvious. The extensive work of Sparrow and his associated (see 22, 25) indicates the importance of the chromosome as one of the sites of the lethal lesion. To simply equate two-hit aberrations to lethality as Puck (10) has suggested, is not valid, as Bender and Wolff (9) had also shown. That loss of reproductive integrity can arise from causes

other than genetic loss at mitosis is also obvious when one considers, for example, the well-known phenomena of interphase death and the work of von Borstel and Reckermeyer (26) which showed the inability of genetic loss to mimic one type of dominant lethality in Habrobracon and Drosophila embryos. Again the mitotic upsets resulting in reproductive sterility reported by Harrington (27) and the inhibition of cell division due to nucleolar inactivation (28) are not obviously related to genetic loss. Thus, the question of the exact role of chromosome aberrations in determining loss of reproductive integrity remains unresolved. The present data certainly indicate that there is no simple relationship between the two events.

SUMMARY

The reproductive integrity of single meristematic cells of Tradescantia occidentalis exposed to acute doses of X rays was investigated. The dose response curve was sigmoid and similar to that reported for a variety of mammalian cell lines having a D_o of 149 r and an extrapolation number of 1.6. Detailed observations were also made of all forms of chromatid and chromosome aberrations induced after irradiating all stages of the mitotic cycle of these same meristematic cells. Attempts were then made to correlate these two sets of data and to equate loss of genetic information to loss of reproductive integrity.

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FOOTNOTES

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[2] I am grateful to Dr. A. H. Sparrow for providing the experimental material.

TABLE I

CHROMATID AND CHROMOSOME ABERRATIONS OBSERVED AT METAPHASE BETWEEN 0 AND 26½ HRS AFTER IRRADIATION

	Fixation-time (Hrs)	Cells scored	Normal	Normal except for gaps	Chromatid gaps	Chromosome gaps	Isochromatid breaks	Chromatid breaks	Chromatid interchanges	Triradials	Chromatid intrachanges	Single minutes	Chromosome breaks	Chromosome exchanges†	Double minutes
36 r	2	50	42	44	3		3	3	2			3			
	4	50	23	40	42		1	4	3			3			
	6	50	20	34	20	1	6	8	6			1			
	8	55	33	42	20	2	6	4	2	1					1
		*(50)	(30)	(38.2)	(18.2)	(1.8)	(5.5)	(3.6)	(1.8)	(0.9)					(0.9)
	10	50	22	31	15	1	7	5	4	1	2	1			
	13⅓	38	22	25	8		8	3	2	1	2		2		2
		(50)	(28.9)	(32.9)	(10.5)		(10.5)	(3.9)	(2.6)	(1.8)	(2.6)		(2.6)		(2.6)
	16	50	36	38	4		7	1			1	3	1		
	18½	50	23	27	10	7	3	7	5		2	8	1		
	21	50	15	16	14	1	11	10	11	1	1	11	1	3	1
	24	50	30	36	9	2	5	1	3				3		
	26½	50	39	39	3	1	3	2				2	5	1	
63 r	2	45	27	33	11	1	6	1	8						
		(50)	(30)	(36.7)	(12.2)	(1.1)	(6.7)	(1.1)	(8.9)						
	4	50	11	25	66	3	7	9	11		5	8		1	
	6	50	3	21	72	4	8	19	4		3	4			1
	8	50	10	20	45	6	13	9	11	2	6	4	1	1	1
	10	49	18	24	13	2	11	7	5	1	6	3		1	
		(50)	(18.4)	(24.5)	(13.2)	(2)	(11.2)	(7.1)	(5.1)	(1)	(6.1)	(3)			
	13⅓	50	22	30	12	1	11	7	4		2		2		
	16	50	20	23	11		14	7	5			5	1	1	
	18½	50	15	23	19		13	6	7		4	6			1
	21	50	22	26	6		16	3	8		1	11	1		
	24	50	18	26	14		10	2	7			6	2	3	
	26½	50	29	33	8		10	2	6		1	2	1	1	
90 r	2	Chromosomes too sticky to be analysable													
	4	50	11	16	59	4	6	14	26	2	3	2			
	6	44	4	8	70	5	18	25	26	3	2	10	1		
		(50)	(4.5)	(9.1)	(77.3)	(5.7)	(20.4)	(28.4)	(29.5)	(3.4)	(2.3)	(11.4)	(1.1)		
	8	28	1	3	46	1	12	17	21	3	7	4			
		(50)	(1.8)	(5.4)	(82.1)	(1.8)	(21.4)	(30.4)	(37.5)	(5.4)	(12.5)	(7.1)			
	10	34	5	11	28	2	14	8	7	4	5	4			5
		(50)	(7.4)	(16.2)	(41.2)	(2.9)	(20.6)	(11.8)	(10.3)	(5.9)	(7.3)	(5.9)			(7.4)
	13⅓	53	16	26	26	3	19	9	14	1	1	2		1	2
		(50)	(15.1)	(24.5)	(24.5)	(2.8)	(17.9)	(8.5)	(13.2)	(0.9)	(0.9)	(1.9)		(0.9)	(1.9)
	16	50	4	12	55	5	16	21	15	4	7	5			
	18½	50	13	22	35	1	16	12	5	4	5	3			1
	21	50	27	34	14		9	10	1			3			
	24	50	22	27	8		10	7	6		1	3	1	4	1
	26½	50	18	22	14		11	13	8	2		3		1	5
Control		100	98	98				2							

* Where the data are based on samples of less than 50 cells, the data have been corrected to aberrations per 50 cells.

† Dicentrics and rings.

TABLE II

EQUATIONS RELATING YIELD OF ABERRATIONS PER CELL (Y)
TO DOSE IN ROENTGENS (D)

Isochromatid aberrations	$Y = .00333\ D$
Chromatid breaks	$Y = .000678\ D + .0000297\ D^2$
Chromatid interchanges	$Y = .000438\ D + .0000319\ D^2$

TABLE III

PREDICTED ABERRATION FREQUENCY PER CELL, AND SURVIVAL, AT HIGHER DOSES

Dose (r)	Chromatid breaks	Isochromatid breaks	Chromatid interchanges	Total chromatid aberr.	% of cells with 0 or 1 aberr.*	Total aberr. excluding chromatid breaks	% of cells with 0 or 1 aberr.*	Observed survival
200	1.336	.666	1.369	3.370	13	2.034	39	40.1%
300	2.903	.999	3.011	6.913	1	4.010	9	18.2%
400	5.071	1.332	5.295	11.698	0.1	6.627	1	10.7%

* A Poisson distribution assumed.

TABLE IVa

PROBABILITY OF SIMULTANEOUSLY DAMAGING TWO HOMOLOGOUS CHROMOSOME ARMS OF *TRADESCANTIA OCCIDENTALIS* (2n = 12)

Number of aberrations	Probability of damaging both homologues
1	0
2	.04
3	.13
4	.25
5	.40
6	.56
7	.71
8	.83
9	.91
10	.96
11	.99

TABLE IVb

Dose (r)	Predicted total chromatid aberr.	Proportion of cells not having any two homologues damaged*	Predicted total chromatid aberr. excluding breaks	Proportion of cells not having any two homologues damaged*	Observed survival
200	3.37	0.77	2.03	0.91	.40
300	6.91	0.36	4.01	0.70	.18

* Poisson distribution of aberrations per cell assumed.

TABLE V

PREDICTED PERCENTAGE OF CELLS WITH 0 OR 1 ABERRATION FROM THE OBSERVED MEAN ABERRATION DOSE*

Dose (r)	Including chromatid breaks M.A.D. = 96 r	Excluding chromatid breaks M.A.D. = 138 r	Observed survival
100	73%	84%	79.8%
200	40%	56%	40.1%
300	20%	35%	18.2%
400	9%	22%	10.7%
500	4.2%	14%	6.1%

* A Poisson distribution of aberrations per cell assumed.

TABLE VI

PERCENTAGE OF CELLS WHICH FROM THEIR METAPHASE CONFIGURATIONS MIGHT BE EXPECTED TO GIVE RISE TO BRIDGES AT ANAPHASE

Dose (r)	% Cells with		
	0 Bridge	1 Bridge	2 or more Bridges
36	86.03	13.05	0.92
63	76.28	19.16	4.56
90	64.49	26.36	9.15

FIGURE LEGEND

Fig. 1. X ray survival curve of terminal hair cells of *Tradescantia occidentalis*.

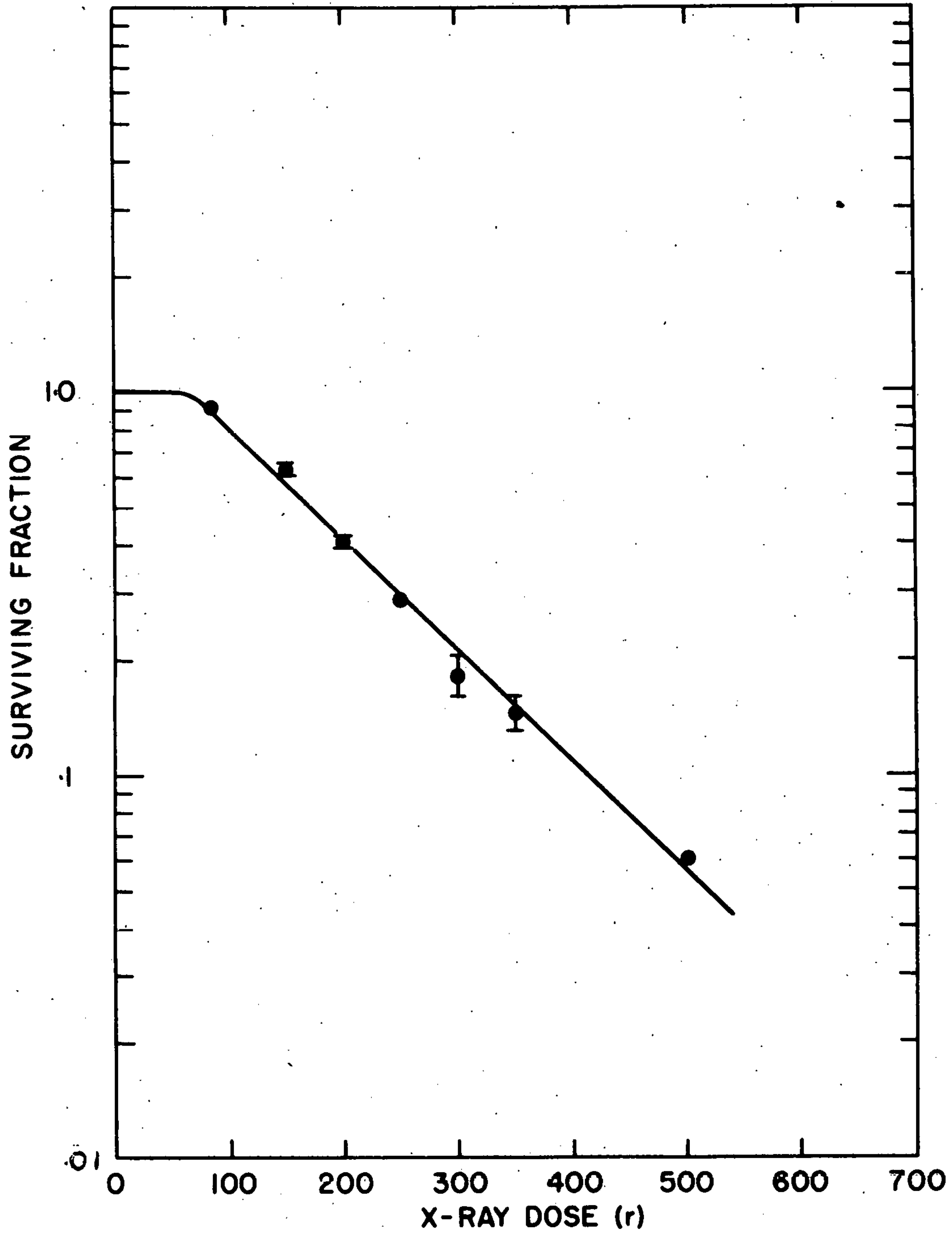

FIGURE 1