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POLLEN ABORTION INDUCED BY GAMMA RADIATION AND ETHANOL IN *Gibasis pulchella**

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SUMMARY

A comparative study of the effects of gamma radiation and ethanol vapors on the development of the male gametophyte in *Gibasis pulchella* was carried out. Dose-response curves were constructed for pollen abortion and for other morphological abnormalities of the pollen grains induced both by gamma radiation an ethanol. The slopes of the curves were similar, but the amplitude of the response was much smaller for the chemical. The sensitive periods in the microsporogenesis cycle were found to be coincident for both agents since the maximum damage was observed between 11 and 15 days after treatment in each case.

In addition to the analysis of pollen abortion, scoring of morphological abnormalities in the pollen grains (double grains and giants) was also carried out. Both agents produced these abnormalities but there were quantitative differences in the response. Furthermore incomplete cytokinesis in pollen mother cells leading to incomplete separation of grains (strangled grains) appeared only in the lot treated with ethanol. These abnormalities might possibly be due to alterations induced during the premeiotic mitosis.

No inhibition of the flowering state was observed for doses lower than 300 Kr, however, doses larger than that produced a decrease in the numbers of flowers as the dose increased and also induced clefting in the petal margins. Ethanol dose of 166 mg/liter of air/3 hr greatly reduced the floral production.

The nuclear volume for the meristematic cells of the root tip of *Gibasis pulchella* is 715.26 $\pm$ 58.29 m^3 and the interphase chromosomal volume is 44.7 m^3. The dose necessary to produce 50% pollen abortion was found to be 332 rads and this value falls on the curve obtained by Underbrink *et al.* (1973).

RESUMEN

Se comparan los efectos producidos por la radiación gamma y los vapores de etanol en el desarrollo del gametofito masculino de *Gibasis pulchella*. Se hicieron curvas de dosis-respuesta para la aborción de polen y para otras anormalidades morfológicas de los granos de polen inducidas por la radiación gamma y por el etanol. Las pendientes de las curvas fueron similares, pero la amplitud de la respuesta fue mucho menor con el agente químico. Los periodos sensibles en

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el ciclo de la microsporogénesis coincidieron para ambos agentes, ya que el daño máximo se observó entre 11 y 15 días después del tratamiento en cada caso.
Además del análisis de aborción de polen también se realizó el registro de anormalidades morfológicas en los granos de polen (granos dobles y gigantes). Ambos agentes produjeron estas anormalidades pero hubo diferencias cuantitativas en la respuesta.
La incompleta citoquinesis en las células madres del polen que condujo también a la separación incompleta de los granos (granos estrangulados) apareció solamente en el grupo tratado con etanol. Estas anormalidades pueden ser posiblemente debidas a alteraciones inducidas durante la mitosis premeiótica.
No se observó inhibición de la floración con dosis menores de 300 kr, sin embargo, dosis mayores a la mencionada produjeron disminución del número de flores a medida que la dosis aumentaba y también indujeron denticiones en las márgenes de los pétalos. Dosis de etanol de 166 mg/litro de aire/3 horas redujo marcadamente la producción de flores.
El volumen nuclear de *Gibasis pulchella* es 715.26 $\pm$ 58.29 m^3 y el volumen cromosómico interfásico es 44.7 m^3. La dosis necesaria para producir el 50% de aborción de polen en 332 rads, este valor puede incorporarse a la curva obtenida por Underbrink *et al.* (1973).

INTRODUCTION

There is a considerable interest in studying the effects of ionizing radiations and chemicals on genetic systems because the damage caused might affect succesive generations. Viability of pollen grains has been a method employed in studying cellular radiation sensitivity in the process of male gametophyte formation. Pollen abortion induced by radiation has been correlated with some nuclear parameters, namely nuclear volume (NV) and interphase chromosome volume (ICV) of different species (Yamakawa y Sparrow, 1966; Underbrink *et al.*, 1973).

Beside the fact that pollen grains have been shown to be sensitive to radiation, they have the advantages of being easy to handle, can be collected in high numbers and the abortion frequency can be determined by means of simple stain techniques (Hauser and Morrison, 1964; Yamakawa and Sparrow, 1966; Alexander, 1969). A very important factor among those influencing radiation sensitivity is the state of microsporogenesis at the time of irradiation. Thus when the treatment is applied, plant material bearing flower buds in various stages of development is the most useful (Yamakawa and Sparrow, 1966; Savage, 1975).

Comparative studies of the action of radiation and chemicals on some clones of *Tradescantia* have been made (Sparrow *et al.*, 1974; Nauman *et al.*, 1974, 1975, 1976).

When applied in liquid form, alcohols such as methanol, ethanol and propanol, have induced chromosomal aberrations in the meristematic cells of *Vicia faba* root tips (Michaelis *et al.*, 1962; Michaelis and Rieger, 1968; Gómez Arroyo and Villalobos-Pietrini; unpublished results).

Furthermore, somatic mutations in the stamen hair system of *Tradescantia* were produced by methanol and ethanol and ethanol in vapor form (Villalobos-Pietrini and Hernández, unpublished results).

The purpose of this work was twofold: first, to compare the response of the pollen grains to exposure to chemicals and to radiation; second, to determine if

pollen abortion is an adequate method to asses the damage induced by the above mentioned agents in order to use it for monitoring of the damage caused by environmental pollutants.

MATERIALS AND METHODS

*Gibasis pulchella,** a perennial herbaceous plant in the Commelinaceae family, was selected because it offers several advantages for these studies such as quick rooting of the cuttings, easy vegetative propagation, fresh flowers blooming daily over long periods of times and a low percentage of spontaneous pollen abortion (4.82%).

1. *Maintenance of plants*

Stock plants were maintained in a greenhouse at the Jardín Botánico Exterior of the Universidad Nacional Autónoma de México. A week before treatment with ethanol or radiation, they were changed to laboratory conditions (25 ± 5°C) and grown in beakers containing aereated Hoagland's solution No. 2 (Conger, 1964) to hasten the rooting.

2. *Irradiation*

Irradiation was carried out in the Hospital General de la Secretaría de Salubridad y Asistencia of Mexico using a ^{60}Co source (Model El Dorado 70). To calibrate the apparatus a Victoreen dosimeter (Model 570) and an ionizing chamber with a rnge of 0— 100 roentgens (Model 621) were used.

Cuttings were placed in a field 26 cm x 26 cm at a distance of 105 cm below the source, with 10 plates of lucite, each 0.5 cm thick over the cuttings and a water phantom under them. Dosimetry studies showed that the intensity of the dosis absorbed was 30.04 rad/min. The doses selected for this study appear in Table I. Depending on the dose, between 5 and 12 cuttings were used.

3. *Chemical exposure*

On the basis of a preliminary work, the concentrations shown in Table III and exposures of 3 hours were selected. For each dose 10 cuttings of *Gibasis pulchella* were used and 5 were kept as controls.

Exposures were made in static closed chambers of 9.0 liters. The cuttings were introduced in them in beakers with 400 ml of Hoagland's solution. The ethanol was placed in a open Petri dish within the chamber and vaporized. No special provision was made to circulate the vapor within the chamber because ethanol is volatile. All of the liquid was vaporized in each exposure (41 to 166 mg/liter).

Quantities of distilled water equal to those of the alcohol were placed in the control chambers.

4. *Pollen viability*

After treatment the cuttings were placed in beakers with fresh Hoagland's solution and mantained at laboratory temperatures 25 ± 5°C and with natural sunlight supplemented by 39 watts General Electric neon lamps throughout the experiment.

Pollen abortion was recorded in flowers blooming daily from the 1st day to the 22nd day after treatment and the mean value for the 5-day scoring period (days 11 to 15 after treatment with radiation or chemicals) was selected because these were the days of the highest frequency of pollen abortion.

Slides of pollen were made following the lactophenol-cotton blue technique described by Yamakawa and Sparrow (1966) and Underbrink *et al.* (1973). Between 200 and 3200 cells were recorded per day per dose.

5. *Nuclear parameters*

The determination of chromosome number as well as nuclear volume was made from meristematic cells of the root tip of the plant.

Meristems were fixed in Craf III, embedded in paraffin and sectioned with a microtome. Slides were stained with safranin and fast green. Two kinds of nuclei were observed: spherical and elongated. The nuclear volumes calculated were based on measurements made on sixty cells with an ocular micrometer using the following relations:

Spherical nuclei: $4/3\ \pi\ r^3$, where r = ½ of the axis length,
Elongated nuclei: $4/3\ \pi\ a\ ^2b$, where a = ½ of the major axis length and
b = ½ of the minor axis length.

Chromosomal number was determined in meristematic cells treated with colchicine and following the acetic-orcein squash technique (Villabos-Pietrini, 1965). Interphase chromosome volume (ICV) was calculated from the mean of the product of the nuclear volumes divided by the number of chromosomes.

RESULTS AND DISCUSSION

Sensitivity of the cells in the different stages of the microsporogenesis.

One of the factors that affects significantly the sensitivity of pollen to mutagens is the state of microsporogenesis at the time of treatment.

The sensitivity/microsporogenesis stage relationship in *Gibasis pulchella* was expressed when daily pollen abortion frequencies exhibited maximal values in flowers

maturing between 11 and 15 days after treatment; hence the most sensitive stages of microsporogenesis occurred 11-15 days before anthesis. If the duration of the different stages of microsporogenesis established in *Tradescantia reflexa* (Sax and Edmonds, 1933) and in *Tradescantia paludosa* (Taylor, 1950) are taken as reference, at the time of treatment, the male gametophyte of *Gibasis pulchella* are between the first and fifth day of microsporogenesis when compared to the cycle of *Tradescantia reflexa* and between the third and seventh day with respect to the cycle of *Tradescantia paludosa*. These days range from premitotic phases to the separation of the tetrad into young microspores. The results obtained in this work are in accord with Beatty and Beatty (1953) who considered the moment in which the central vacuole is spreading, after the separation of the teatrad into independently growing microspores as the most sensitive stage to physical and chemical agents. The results obtained by Yamakawa and Sparrow (1966) support these observations and establish that the most sensitive period in the development of the male gametophyte is related to the meiotic phases; that is, from the first to the sixth day in the development of the pollen grains in the different species of *Tradescantia*.

As it has been established that the duration of microsporogenesis is not very different among the various genera of the Commelinaceae (Sax and Edmonds, 1933; Maheshwari, 1949; Taylor, 1950; Beatty and Beatty, 1953) an extrapolation can be made for *Gibasis pulchella*. Thus the flowers that opened between the eleventh and the fifteenth day were assumed to be in the meiotic state when they were treated. Therefore, these buds that were most sensitive had been treated as early as between the first and the third day before the meiotic prophase, during the sporogenic stages when the pollen mother cells were in the differentiation process (Taylor, 1950). These results agree with those obtained by Yamakawa and Sparrow (1966), who concluded from their cytological investigations of three species that the development of the pollen mother cells begins fifteen or sixteen days before the blooming, which is coincident with an increase of pollen abortion.

Dose-response curves

The peak period of pollen abortion was between the eleventh and fifteenth day after treatment for all of the doses used. The data from these days were grouped in order to compare our results with those of Underbrink *et al.* (1973). We also ascertained the maximal value from this period (Table I). In all cases the control value of 4.82% has been subtracted. Doses of 10, 20 and 30 rads (Table I) produce increases in pollen abortion but to levels which do not exceed the 95% confidence limits of a doubled background abortion frequency.

By means of a X^2 test it was determined that the smallest dispersion of the values obtained with respect to the values expected, corresponded to a geometric function.

TABLE I

POLLEN ABORTION AFTER ^{60}CO GAMMA IRRADIATION IN *GIBASIS PULCHELLA*

Dose (rads)	*Length of Exposure (min)*	*Grouped data 11-15 days postirradiation (minus control)*	*Maximal value (minus control)*
		% ± S.E.	% ± S.E.
10	0.33	2.364 ± 0.41	2.654 ± 0.63
20	0.66	2.817 ± 0.66	5.275 ± 0.75
30	1.00	3.748 ± 1.03	6.403 ± 0.66
50	1.66	12.223 ± 2.13	19.647 ± 1.99
100	3.33	19.761 ± 1.78	28.647 ± 1.74
200	6.66	36.337 ± 5.99	49.451 ± 0.86
300	10.00	38.617 ± 3.13	47.426 ± 1.39
400	13.33	31.061 ± 7.69	52.573 ± 0.44
500	16.66	40.124 ± 5.45	54.452 ± 3.63
600	20.00	56.299 ± 7.59	78.734 ± 8.11
700	23.33	54.414 ± 4.16	61.300 ± 3.82
800	26.66	66.716 ± 3.26	74.504 ± 2.57
900	30.00	73.956 ± 2.18	80.119 ± 4.46
1000	33.33	76.004 ± 3 04	84.738 ± 2.36
1200	40.00	73.372 ± 3.51	81.071*

* Data from only one experiment

The relations obtained from the regression lines by the method of least squeares are the following:

i) for maximum values:

$$\log Y = 0.572 + 0.447 \log X$$

ii) for grouped data:

$$\log Y = 0.158 + 0.563 \log X$$

where X = gamma dose in rads

Y = percent of pollen abortion

These equations and the correlation coefficients are shown in Table III to facilitate the comparison of grouped data and maximum values.

TABLE II

VALUES OF REGRESSION LINES OF DOSE-RESPONSE CURVES FOR GROUPED DATA AND FOR MAXIMAL PERCENTAGE OF POLLEN ABORTION AFTER GAMMAL IRRADIATION

	Intercept	*Slope*	X^2	*Correlation coefficient*
Grouped data	0.158	0.563	8.913*	0.967
Maximum value	0.572	0.447	7.738*	0.971

* Not significant, $X^2_{0.95\ (11)} = 19.7$

Dose-response curves in Fig. 1 compare the grouped data corresponding to the peak days with data for the maximum single-day percentaje of pollen abortion independent of the day on which it occurred, as described by Underbring *et al.* (1973). Both methods demonstrate that pollen abortion increases linearly within corresponding dose increase. In addition, the slopes obtained from the regression lines for the grouped data and for the maximum values are so similar that they are indistinguishable. However, using the X^2 goodness of fit test one sees that the smaller dispersion and the better correlation correspond to the data for maximum frequencies of pollen abortion, coinciding with the method used by Underbrink *et al.* (1973). Dose to produce 50% pollen abortion in the values of maximum percent of pollen abortion is 332 rads, the dose-response curve in a log-log plot has a slope of +0.447 for the maximum values which is significantly less than one; although this type of curve is difficult to explain; for Underbrink *et al.* (1973) the effect to flatten the slope could be due to the deposition of an excess of energy. As can be observed in Fig. 1, that from 900 rads on, the degree of polen abortion does not increase significantly; thus, we decided to fit the line only up to 900 rads. The regression lines for the maximum daily percentage in both cases are very similar:

i) for all the doses:

$$\log Y = 0.572 + 0.447 \log X$$

ii) for doses up to 800 rads:

$$\log Y = 0.554 + 0.455 \log X$$

Considering maximum values, the saturation point is reached at 900 rads. Yamakawa and Sparrow (1966) observed that increasing the dose in the saturation range produced a marked reduction in the number of grains in the anthers but dit not change the percentage of grains aborted.

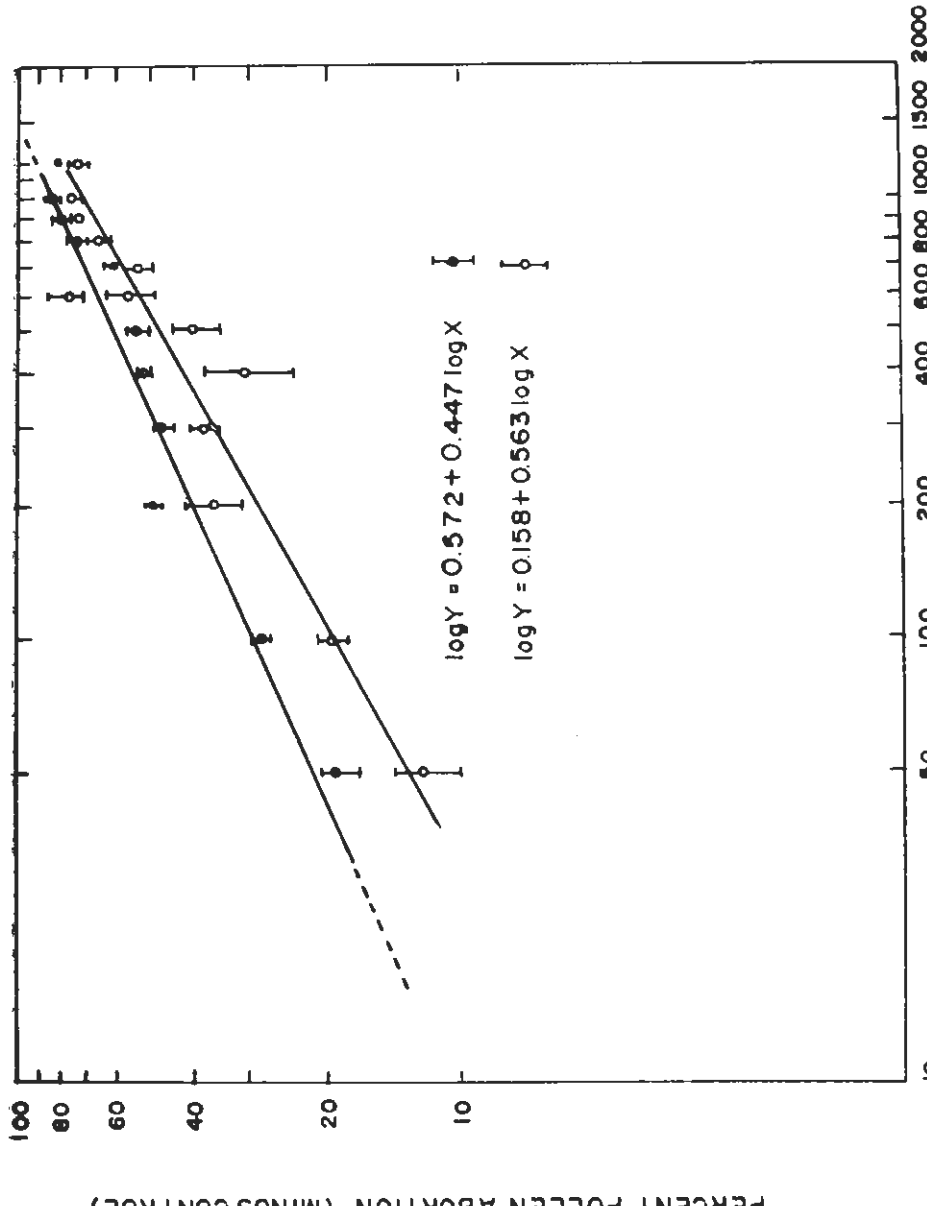

Fig. 1. Dose-response curves for pollen abortion following gamma irradiation (minus control). (○): based on data collected during a 5-day scoring period (days 11-15) and, (●): maximum percentage for *Gibasis pulchella* ± S.E

In the case of treatment with ethanol, the sensitive period coincided with that obtained with gamma irradiation, that is, between days eleven fifteen after treatment. Table III presents the frequencies of pollen abortion for the grouped data. The single day maximum values presented such a wide variability that it was not considered advantageous to include them in the analysis. The relation which best fits the data was the geometric function with the following equation:

$$\log Y = (-0.299) + 0.683 \log X$$

TABLE III

POLLEN ABORTION AFTER TREATMENT WITH ETHANOL IN *GIBASIS PULCHELLA*

Dose mg/liter (3 hr exposure in air)	*Grouped data 11-15 days after treatment (minus control)*
	% ± S.E.
41.47	6.15 ± 2.60
62.21	8.52 ± 0.50
82.95	11.89 ± 4.62
103.68	12.28 ± 4.32
124.42	10.71 ± 2.97
145.16	14.73 ± 3.08
165.89	18.41 ± 2.92

In Fig. 2 the data are presented graphically and it can be observed that the percentage of pollen abortion increased as the dosis of ethanol increase. However, if the results obtained for both agents are compared, arbitrarily setting 1 mg/liter = 1 rad, one notes that the range of pollen abortion produced by the different percentages of ethanol vapors is limited by the experimental design (an increase of only 6% to 18% over controls), whereas the design for gamma radiation produced a wide response. The equivalence used is arbitrary and is introduced to compare the results obtained both for ethanol vapors and for gamma radiation on the same set of axes.

Koller (1943) has indicated that the frequency of chromosomal aberrations in the pollen grains of *Tradescantia* increases from 16% with 50 roentgens of gamma radiation to 71% with 200 roentgens. The induction of aborted pollen grains and in some cases of cell death can be attributed to the induction of chromosomal aberrations in the sensitive stages of microsporogenesis both by gamma radiation and ethanol vapors. Vapors of some other chemicals such as methyl chloride, ethylene oxide and ketene have been shown to induce chromosomal aberrations in the pollen tube of *Tradescantia paludosa* (Smith and Lofty, 1954). Ethanol applied in liquid form also induces chromosomal aberrations in the meristems of the root tips of *Vicia faba* (Michaelis *et al.*, 1962; Gómez-Arroyo and Villalobos-Pietrini, unpublished results).

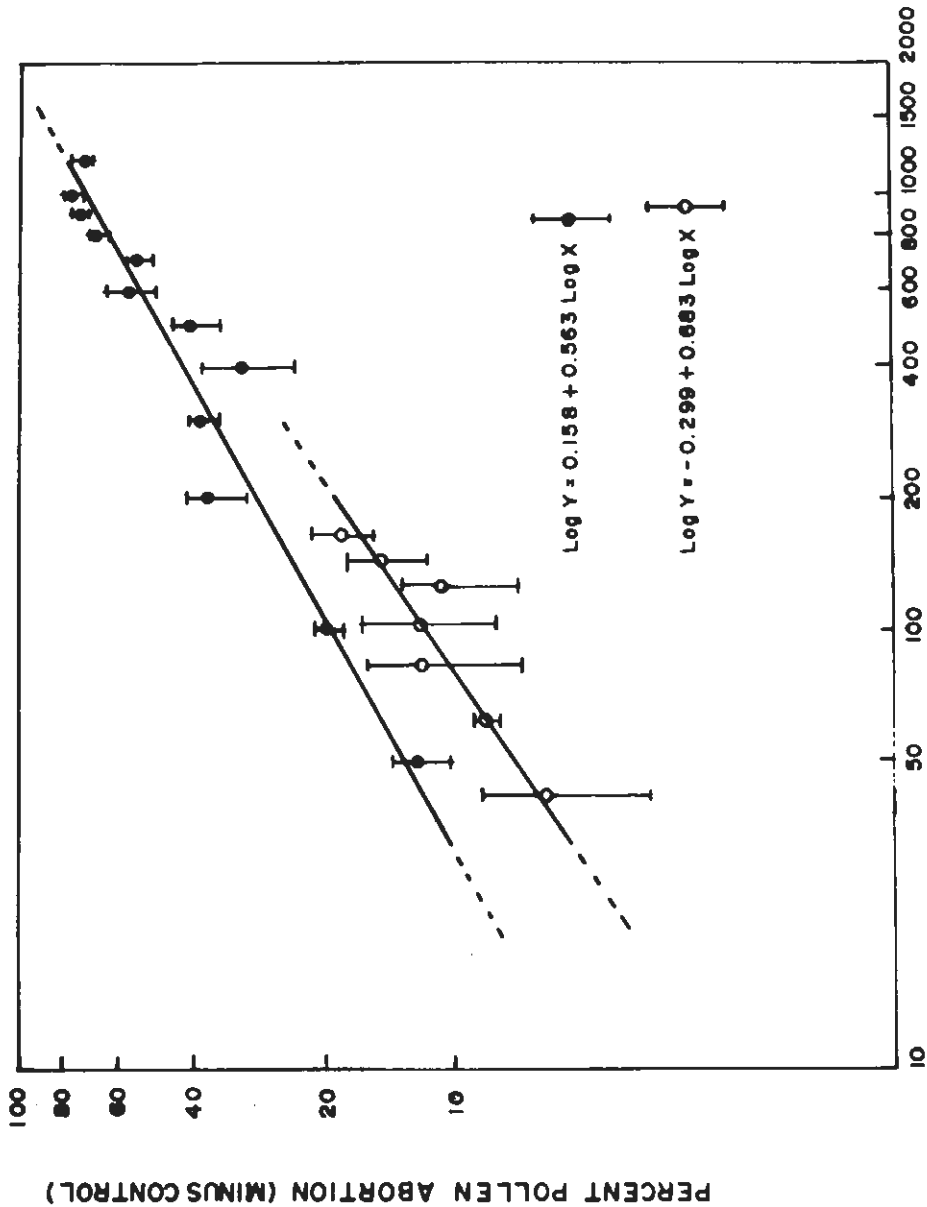

Fig. 2. Dose-response curves for pollen abortion after exposure to ethanol and gamma irradiation for *Gibasis pulchella*. (●): Dose in rads form gamma irradiation ± S.E. and (○): in mg/liter/3 hr for ethanol ± S.E.

Abnormal cells

In slides of pollen grains from plants treated with gamma radiations and ethanol besides abortion pollen grains, some morphological abnormalities were observed, abnormal forms as double grains and grains with a greater than normal volume. The normal volume was 22,640 + 331 μ^3 and larger grains were divided into two groups: large with a mean of 43,660 + 304 μ^3 and giant with a mean volume of 96,100 $\pm$ 313 μ^3. These abnormalities are induced by all doses higher than 200 rads of gamma rays (Table IV) and by all of the doses of ethanol (Table V). Ethanol vapors also induced a morphological effect we term strangled grains, which were also observed. This effect was not observed in irradiated material.

TABLE IV

ABNORMAL GRAIN FREQUENCY IN *GIBASIS PULCHELLA* AFTER GAMMA IRRADIATION

Dose (rads)	*Double grains*	*Large grains*	*Giant grains*	*Total of abnormal grains*
300	0.0008	0.0006	0.0004	0.0018
400	0.0009	0.0006	0.0008	0.0023
500	0.0018	0.0026	0.0021	0.0065
600	0.0026	0.0052	0.0019	0.0097
700	0.0008	0.0046	0.0013	0.0067
800	0	0.0025	0	0.0025
900	0.0063	0.0241	0.0133	0.0437
1000	0.0019	0.0096	0.0019	0.0134
1200	0	0.0200	0.0100	0.0300

TABLE V

ABNORMAL GRAIN FREQUENCY FOR *GIBASIS PULCHELLA* AFTER TREATMENT WITH ETHANOL

Dose mg/liter (3 hrs exposures in air)	*Double grains*	*Large grains*	*Giant grains*	*Strangled grains*	*Total of abnormal grains*
41.47	0.0001	0.0017	0	0.0001	0.0019
62.21	0	0.0020	0	0	0.0020
82.95	0	0.0011	0	0	0.0011
103.68	0.0009	0.0010	0	0.0064	0.0083
124.42	0.0009	0.0124	0.0032	0.0169	0.0334
145.16	0.0006	0.0375	0.0035	0.0021	0.0437

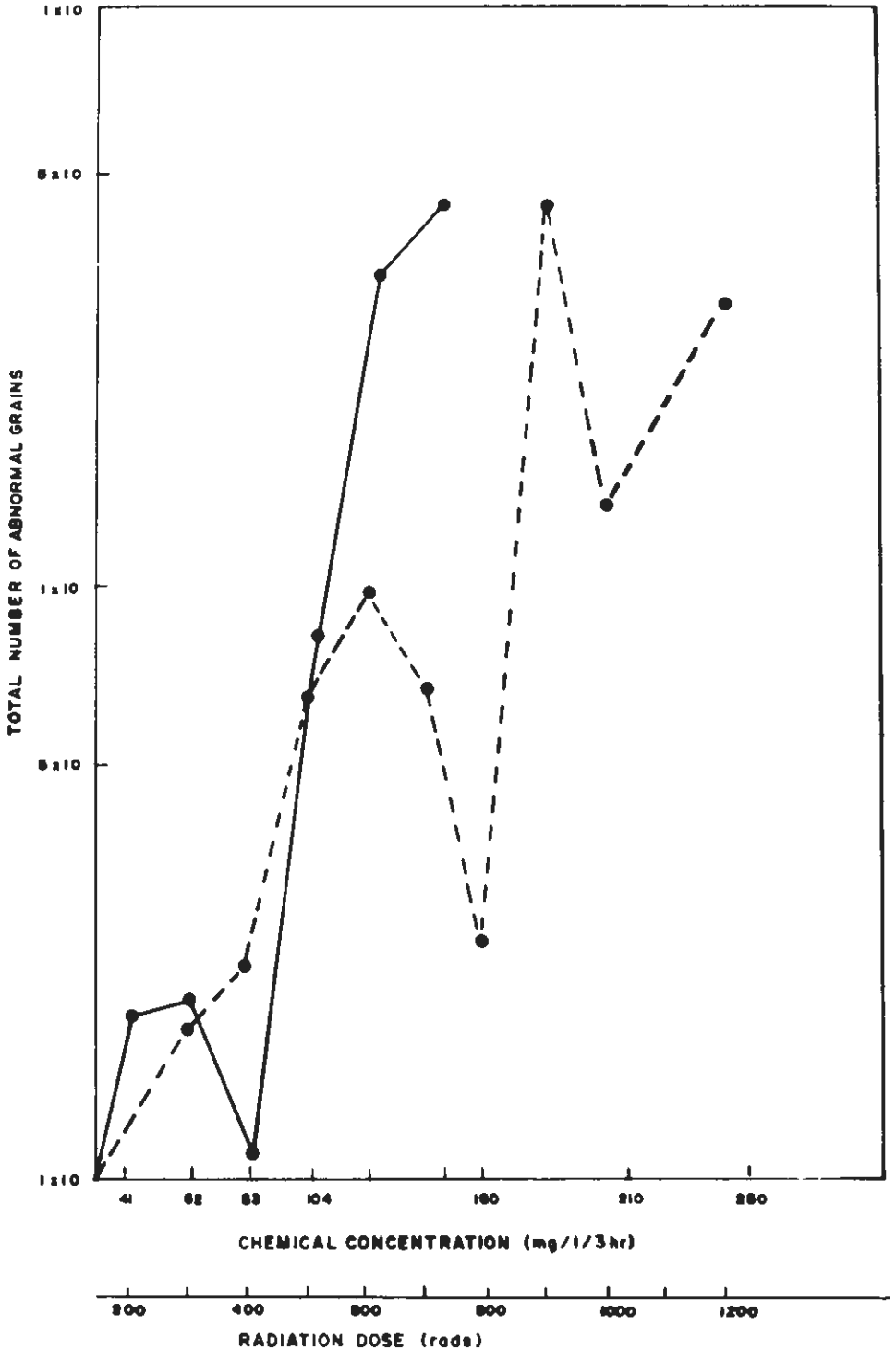

Fig. 3 Dose-response curves for abnormal grain induction after exposure to ethanol (—) and gamma irradiation (— — —).

In Fig. 3, 1 mg/liter/3 hr was arbitrarily set equal to 5 rads and the frequencies of abnormal grains produced by both agents are shown. In general there is a tendency for the number of these cellular types to increase as the dose increases. Probably the induction of abnormal grains will prove to be a useful method for measuring the effect caused by ethanol and perhaps for some environmental contaminants.

Binucleate or multinucleate pollen mother cells larger than normal ones, are produced from pollen grains of large volume. These abnormalities appear spontaneously and in some cases may be the effect of viral infections (Swaminathan *et al.*, 1959) or may be caused by exposure to X rays or neutrons (Kamra, 1960). However, it was found that there were no significant differences between the spontaneous frequency and that produced by these mutagenic agents (Kamra, 1960). In all cases the frequency is very low, namely, 1.2% binucleate cells in an oat hybrid (Holden and Mota, 1956), 2.4% binucleate and 0.2% multinucleate cells in *Hordeum* (Kamra, 1960). These studies postulate that the binucleate condition could be the product of a defect in the formation of the cellular wall in the pollen mother cells or in the mitotic spindle during the premitotic mitosis, which expreses itself as an increase in the genom and in the pollen grain volume.

TABLE VI

FLOWER PRODUCTION IN *GIBASIS PULCHELLA* AFTER GAMMA IRRADIATION (11-15 DAYS, GROUPED DATA)

Dose	*% Flowering (related to the control)*
10	96
20	93
30	96
50	100
100	100
200	98
300	100
400	74
500	42
600	38
700	38
800	12
900	11
1000	10
1200	4

Floral production

There were also morphological and numerical differences in the floral production between the material treated with gamma radiation and the control from the eleventh to the fifteenth day after irradiation. No inhibition of flowering was observed for doses of 300 rads or lower (Table VI); however, larger doses than

300 rads produce a decrease in the number of flowers as the dose increases. Flower production by ethanol exposures is in Table VII. Ethanol exposures of 165.89 mg/liter/3 hr reduced the floral production by 45% compared to the control, at this dose the chamber was saturated. When the exposure was larger (6 hours), floral inhibition occurred.

The floral morphology was affected, too. Doses larger than 300 rads induce notching in the petal borders of the flowers that open from the twelveth to the twentieth day after irradiation. Moreover, there was a reduction in size of all floral structures. Comparing these effects with the percentage of pollen abortion one can note that floral parts of flowers produced following exposures to radiations seemed less affected than the pollen. This agrees with the work of Yamakawa and Sparrow (1966) in *Nigella* and *Arabidopsis* where the dose that produced 50% of pollen abortion did not affect floral organs. These authors concluded that in *Nigella*, *Tradescantia* and *Lycopersicum*, there are two periods sensitive to radiation: one of them corresponds to the formation of pollen, and the other to the differentiation and growth of the floral organs. In *Gibasis pulchella* both periods coincide since the peak days in the two cases were from the eleventh to the fifteenth day, and notching in the petal borders was observed from the twelfth day after treatment. This could be a result of the active somatic cell division in petals and the formation of the pollen grains occuring simultaneously.

TABLE VII

FLOWER PRODUCTION IN *GIBASIS PULCHELLA* AFTER GAMMA IRRADIATION (11-15 DAYS, GROUPED DATA)

Dose mg/liter (3 hr exposure in air)	*% lowering (related to the control)*
41.47	82
62.21	71
82.95	80
103.68	63
124.42	51
145.16	56
165.89	45
165.89/6 h	inhibition of flowering

Prediction of sensitivity

The chromosomal number for *Gibasis pulchella* was 2n = 16; six pairs of chromosomes are metacentric and two pairs are acrocentric. In the latter, the nucleolar organizers were observed. The nuclear volume (NV) obtained was 715.26 ± 58.29 m^3 and from these data the interphase chromosomal volume (ICV) = 44.7 m^3 was calculated.

Within the *Gibasis* genera the species geniculata (Jacq.) (Rohw. clone 2235) is the most similar to *pulchella* in its nuclear characteristics with NV = 670.5 m^3, ICV = 41.9 m^3 and 2n = 16 (Sparrow, Schairer and Nauman, personal communication). The spontaneous pollen abortion in *Gibasis geniculata* is 8.5% (Sparrow, Schairer and Nauman, personal communication) while for *Gibasis pulchella* it is 4.82%.

TABLE VIII

NUCLEAR CHARACTERISTICS AND DOSES TO PRODUCE 50% POLLEN ABORTION DETERMINED FROM DOSE-RESPONSE CURVES (FROM UNDERBRINK *ET AL.*, 1973) INCLUDING *Gibasis pulchella*

Species	*Species code*	*Chromosome number (2n)*	*Ploidy level (x)*	*Nuclear volume (μ^3)*	*I C V (μ^3)*	*50% Pollen abortion dose (rads)*
Floxcopa scandens	1	54	6	538.7	10.0	800
Gibasis geniculata	2	16	2	670.5	41.9	105
G. karwinskyana (1974)	3	20	4	913.6	45.7	105
Tradescantia sp. (2084)	5	29	c.2	918.0	31.6	265
T. blossfeldiana	6	72	12	504.0	7.0	600
T. commelinoides	7	16	2	621.0	38.8	225
T. crassula	8	72	12	506.0	6.8	490
T. paludosa (B2-2)	9	12	2	787.1	65.6	90
T. paludosa (2465)	10	12	2	905.8	75.5	76
T. subacaulis	12	12	2	664.5	55.4	88
T. virginiana	13	24	4	1381.0	57.5	90
Tripogandra elongata	14	64	8	488.9	7.7	700
Gibasis pulchella	16	16	2	715.3	44.7	330

Earlier investigations have shown that an inverse relation exists between NV, ICV and DNA content/cell and the radiosensitivity of the material. These studies were done using various criteria for sensitivity, such as growth inhibition (Sparrow *et al.*, 1963), letal doses in some points of the survival curve (Cabrero and Villalobos-Pietrini, 1968; Underbrink *et al.* 1968; Sparrow and Schwemmer, 1974), pollen abortion (Yamakawa and Sparrow, 1966; Underbrink *et al.* 1973; Underbrink and Pond, 1976), etc. In most of the cases the relation is logarithmic and the slope is close to —1. From these curves one can predict the species radiosensitivity, knowing their nuclear volume and their interphase chromosome volume, or DNA content per cell.

The dose required to produce 50% pollen abortion in *Gibasis pulchella* (330 rads) is plotted on the graph obtained by Underbrink *et al.*, where they establish the doses necessary for the induction of 50 per cent pollen abortion versus the ICV and NV (Figs. 4 and 5, Table VIII).

Their values are based on measurements from vegetative shoots, and ours are based on root tip nuclei. Underbrink *et al.* (1973) has reported, in the case of ICV, a correlation coefficient of —0.947. This value changes to —0.904 when Gibasis data is added.

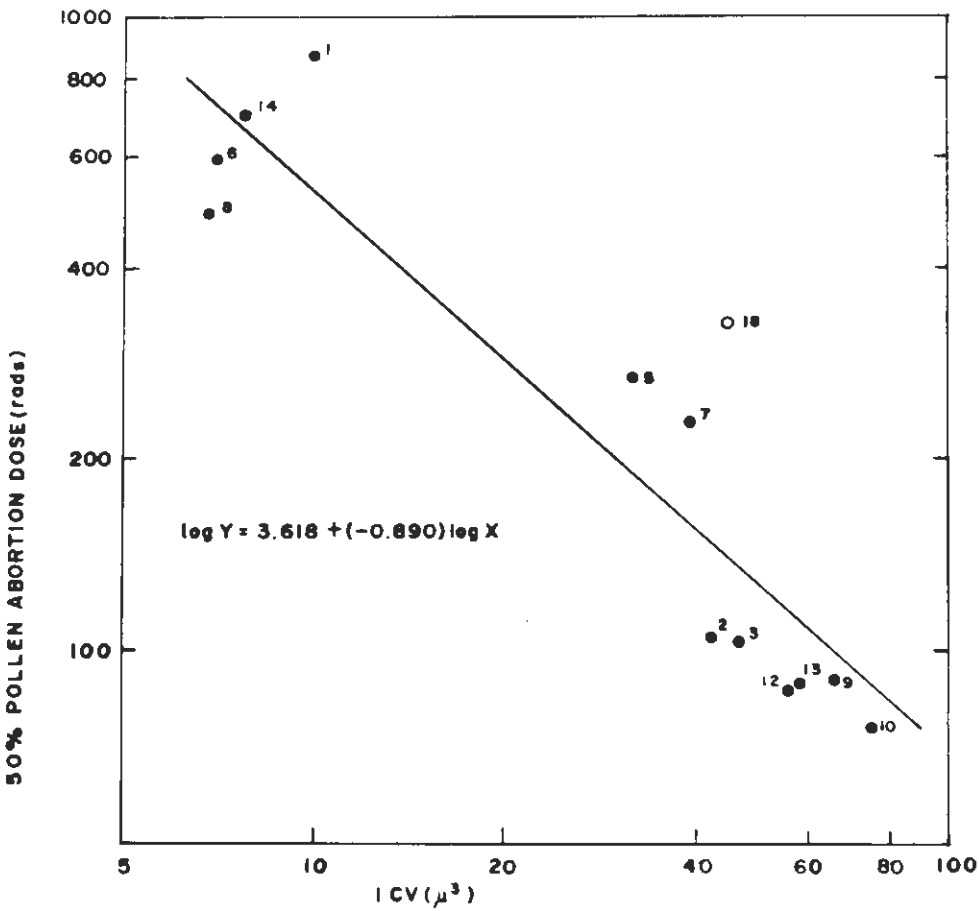

Fig. 4. Doses required to produce 50% pollen abortion following low LET irradiation vs interphase chromosome volume (ICV) for several species including *Gibasis pulchella* (◯).

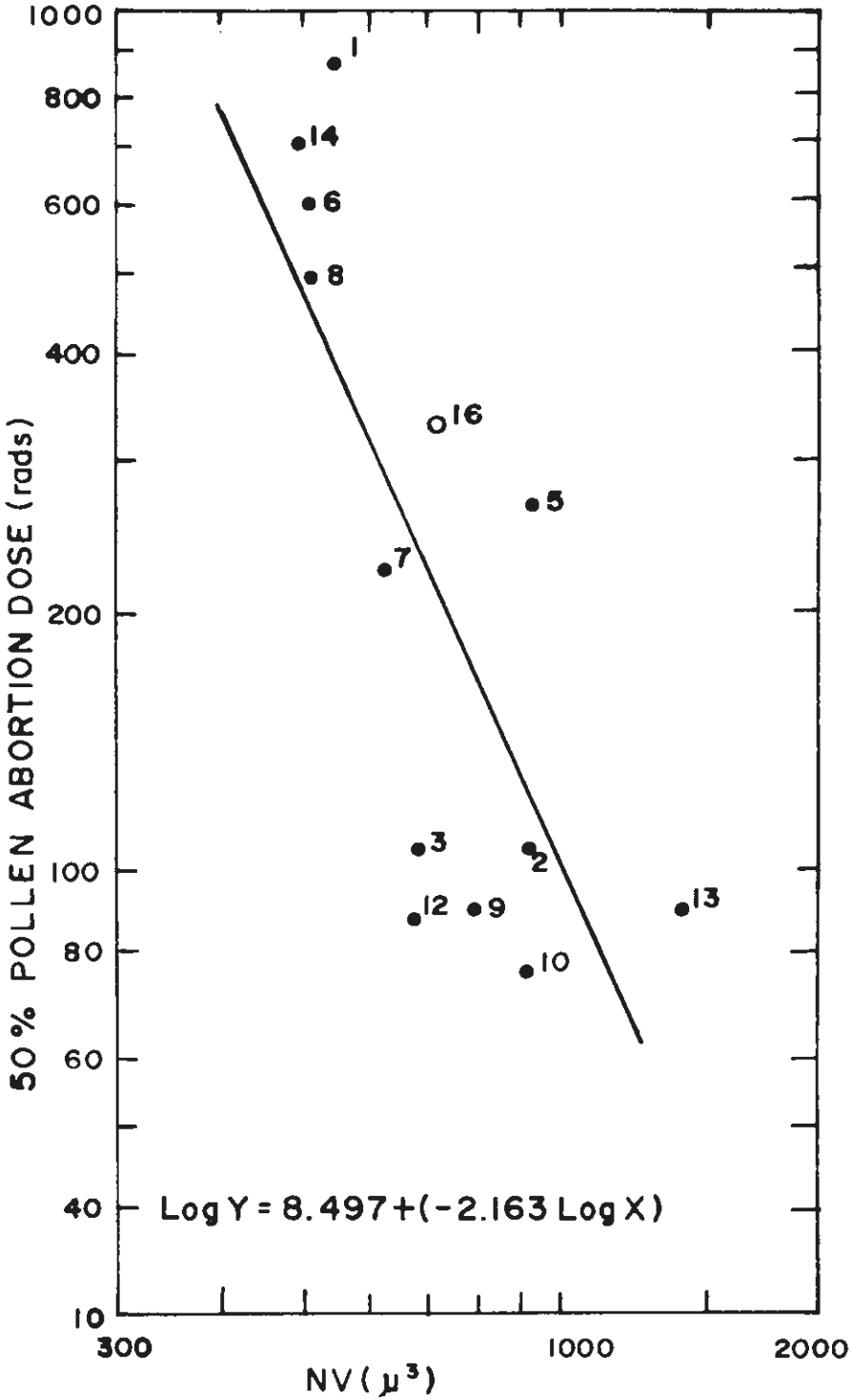

Fig. 5. Doses required to produce 50% pollen abortion following low LET irradiation vs nuclear volumes (NV) for several species including *Gibasis pulchella* (◯).

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