

THE EFFECTS OF STORAGE ON CO⁶⁰-GAMMA RADIATED VICIA FABA. SEEDS

by

HODA HAKEEM

Univ. College of Girls,

Ein-Shams University, Helipolis, Cairo, U.A.R.

The use of radiation in practical breeding research has attracted considerable attention during the last years. In this respect, efforts were much directed towards general radiation effects on plants and seeds 1, 5 & 6. Although much work had been done on the nature and mechanism of chromosome damage by ionising radiation, yet further studies concerning analysis of chromosome breakage and the nature of chromosome aberrations seemed necessary.

The present work is concerned with the effect of Co⁶⁰ gamma radiation on mitosis of *Vicia faba*. The effect on root mitosis of stored radiated seeds had been also studied.

MATERIAL AND METHODS

Seeds of *Vicia faba* (Var. Rebaya 40) were irradiated by Co⁶⁰-gamma rays. Irradiation doses were 4000 r, 7000 r, 12000 r, 16000 r, 20000 r and 25000 r, 100 irradiated seeds were planted in sawdust ; 100 untreated seeds were cultivated under the same experimental conditions and were used as controls. Half of the irradiated seeds were planted 48 hours after irradiation, the other after 4 months from the date of irradiation.

For cytological studies, the root tips of 50 seedlings from each treatment were taken. 30 roots were stained using the acetoorcein squash technique. The other 20 roots were prepared by the paraffin method. Longitudinal as well as cross sections (10 micron thick) were studied after staining with gentian violet.

RESULTS AND DISCUSSION

Study of root cells of unstored and stored irradiated seeds showed that the mitotic rate was, as expected more affected by the higher doses. The rate of cell division decreased in the 20,000 r and 25,000 r (Table 1

and 2). In the lower doses, the rate of mitosis was more or less the same as in the control.

The percentage of induced abnormalities increased with increased doses of irradiation. The types of abnormalities differed in unstored irradiated seeds from that in stored ones. Root cells from unstored irradiated seeds showed more structural abnormalities than physiological such as breaks and micronuclei. (Table 1). Root cells from stored irradiated seeds showed more or less physiological than structural ones. (Table 2).

Stickiness of the chromosomes in metaphases and ana-telophases was observed. Lower doses affected light stickiness of the chromosomes, whose percentage was relatively high (Table 2). The degree of stickiness reached its maximum in the higher doses. In the metaphases stickiness varied from light stickiness to the stickiness of the chromosomes in one or more clumps. (Fig. 1-3). In the ana-telophases the result was the occurrence of sticky bridges Fig. 4-5.

Stickiness was reported to occur as a result of irradiation with X-rays (Sax 1941), or as a result of treatment with various agents. e. g. Podophyllin (Hussain and Hakeem 1961 ; Amer and Hakeem 1964), Stickiness induced by X-ray-irradiation was attributed to excess charge of nucleic acid on the chromosomes (Sax 1941).

The disturbances in the anaphases ranged from one or more (Fig. 5) lagging chromosomes to the spreading and irregular scattering of all the chromosomes all over the cell (Fig. 6). The latter type of disturbance might be attributed to complete disturbance of the spindle. Another type of abnormalities in the anaphases was the anaphase-groups : in which the chromatids were present in groups but not in true poles. Such anaphase type may originate from the metaphase clumps of chromosomes by longitudinal splitting of the chromosomes. Because of stickiness the chromosomes may be unable to move to different poles and thus lead to the above mentioned abnormality. Such anaphase type was also observed as a result of treatment with podophyllin (Amer & Hakeem 1964). Micronuclei and multipolar anaphases were also observed as a result of irradiation with all the employed doses. Tripolar anaphases were the most prominent (Fig. 7). Garcia (1960) treating *Vicia* plants with Co^{60} gamma radiation found, micronuclei, agglutination and fragmentation of chromosomes.

The percentage of chromosome breakage was relatively small (Table 1). This may be due to stickiness which hinders the appearance of chromosome breaks.

TABLE I

Rate of cell division and percentages of abnormalities as effected by Co^{60} gamma radiation. (unstored seeds).

Dose (kr)	No. of cells counted	No. of divid. cells	% of cell divis.	% of abnorm cells	Percent. of the different abnormalities					
					multipl. & anaph. with lag. chromos. micro nuclei	Fully disturb anaphases	Loss of chrom. activity	Breakage	Stickiness	
Cont.	5000	317	6.34	—	—	—	—	—	—	—
4	5000	255	6.1	10.2	31.1	18.5	11.3	38.2	17	—
7	5000	254	5.4	12.5	10.3	25.1	14.9	27.6	12.2	—
12	5000	298	4.96	12.9	21.6	16.4	7	28.6	18	—
16	5000	219	3.98	19.8	21.1	26.5	7.1	38.1	11	—
20	5000	212	3.24	23.9	20.1	26.6	17.6	27.2	8.5	—
25	5000	204	3.08	25.1	23.6	11.1	11.1	18.1	9.7	—

TABLE 2
Rate of cell division and percentages of abnormalities as affected by Co⁶⁰ gamma radiation. (Stored seeds).

Dose (kr)	No. of cells counted	No. of divid. cells	% of cell divis.	% of abnorm cells	Percent. of the different abnormalities					
					multipol. anaph. with lag. chromos. micro nuclei	Fully disturb anaphases	Loss of chrom. activity	Breakage	Stickiness	
Cont.	5000	310	6	—	—	—	—	—	—	—
4	5000	290	5.98	11.0	34.1	10.5	11.5	16.1	38	
7	5000	229	4.6	13.6	30.3	15.1	14.9	9.5	32	
12	5000	251	5.01	15.1	31.5	16.4	11	8.1	26	
16	5000	199	4.00	20.1	26.4	21.5	11.1	8.0	30	
20	5000	200	4.00	21.0	22	25.9	16.5	7.1	28.6	
25	5000	161	3.21	23.9	15.3	32.6	18.0	7.0	2.5	

In addition to stickiness, loss of chromaticity (Fig. 8-9) was frequently met with, especially in higher doses of irradiation. Sometimes it confuses with breakage of chromosomes, yet keen examination can differentiate the two abnormalities. Thus in stored seeds Co^{60} gamma radiation seemed to affect more the chemical chromosome structure and nucleic acid metabolism, giving rise to the above abnormalities.

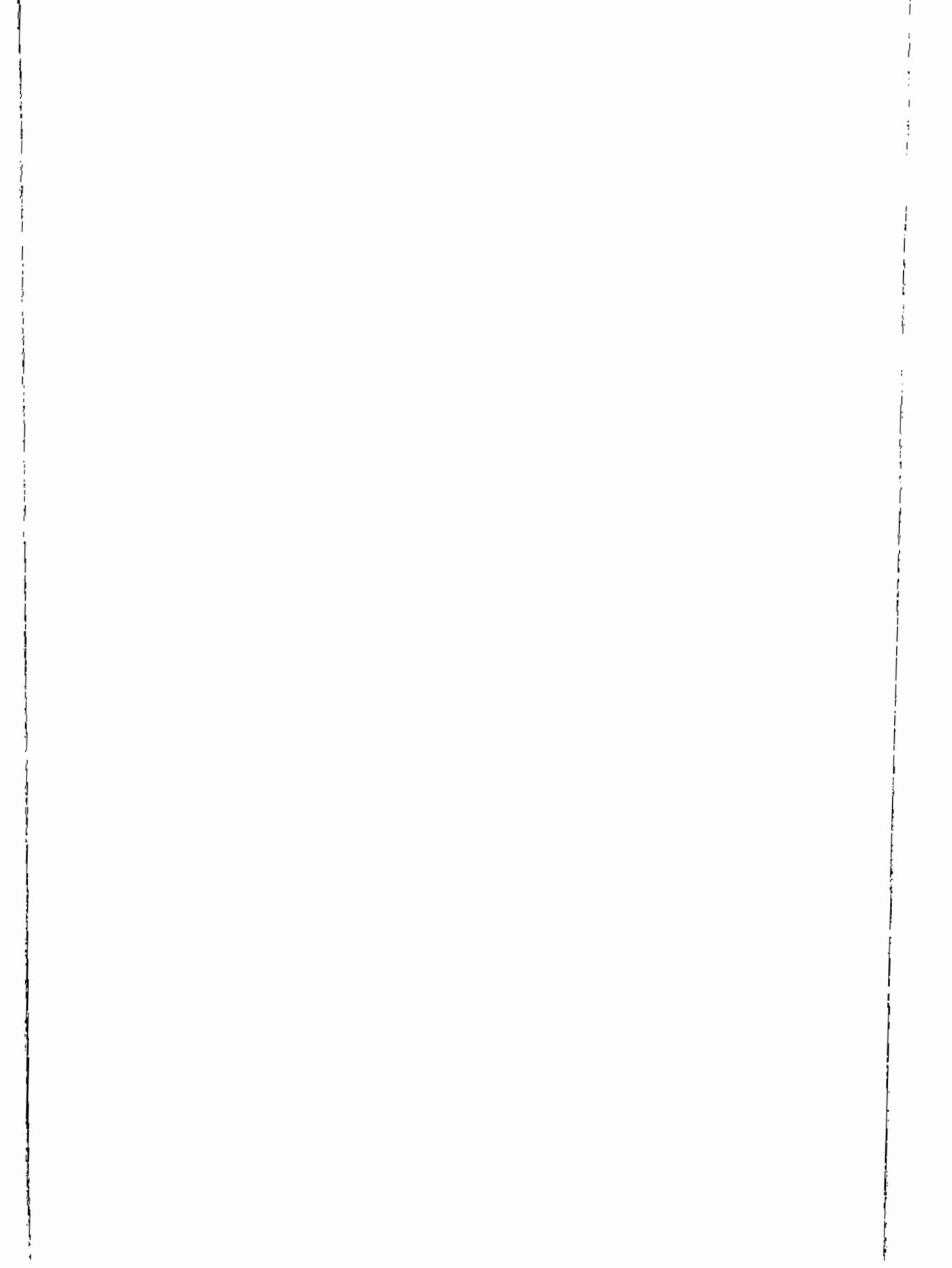
SUMMARY

Dry seeds of *Vicia faba* (Var. Rebaya 40) were stored for 4 months after irradiation comparing unstored irradiated seeds with stored ones :

1. Rate of mitosis was not affected by storage period.
2. Percentage of abnormal cells was not affected.
3. Types of abnormalities deviated to physiological ones, rather than structural ones, in stored irradiated seeds.

REFERENCES

1. Amer, S. and H. A. Hakeem (1964) The effect of Co^{60} gamma radiation on *Lupinus Termis* Rad. Bot. 4 : 95-100.
2. Amer, S. and H. Hakeem (1964) Über die merokinetische Wirkung de Podophyllins. Beitr, Biol, Pflanzler. 40 : 283-292.
3. Garcia M. H. (1960) Ensayos sobre la accion del Co^{60} en la division celular de *Vicia faba*. Genet. iber. 12 : 53-97. (Plant breeding abst. 31 : 18 (1961).
4. Hussein, F. and H. Hakeem (1961) Cytological effects of podophyllin on *Vicia faba* and *Luffa cylindrica*. U.A.R. J. Bot., 111 : 85-98.
5. Karlaruhe Symposium (1961). (IAEA-FAO STI pnb 113).
6. Neary, G. J., S. N. Tonkinson and F. S. Williamson (1959). The relative biological efficiency of single doses of fast Neutrons and gamma-rays on *Vicia faba* roots and with effect of oxygen. I Reduction of root growth. Iter. J. Rad. Biology. 4 : 201-215.
7. Sax, K. (1941) The behaviour of X-ray induced chromosomal aberration in *Allium* root tip cells. Genetics, 26 : 418-425.



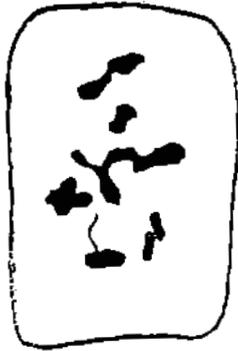


Fig. 1



Fig. 2

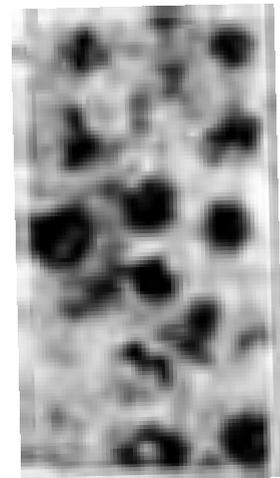


Fig. 3

Fig. 1-3. Different type of sticky metaphases induced by irradiation with $^{16}\text{K}_\alpha$ (Fig. 1) and ^{25}Kr (Figs. 2 & 3).

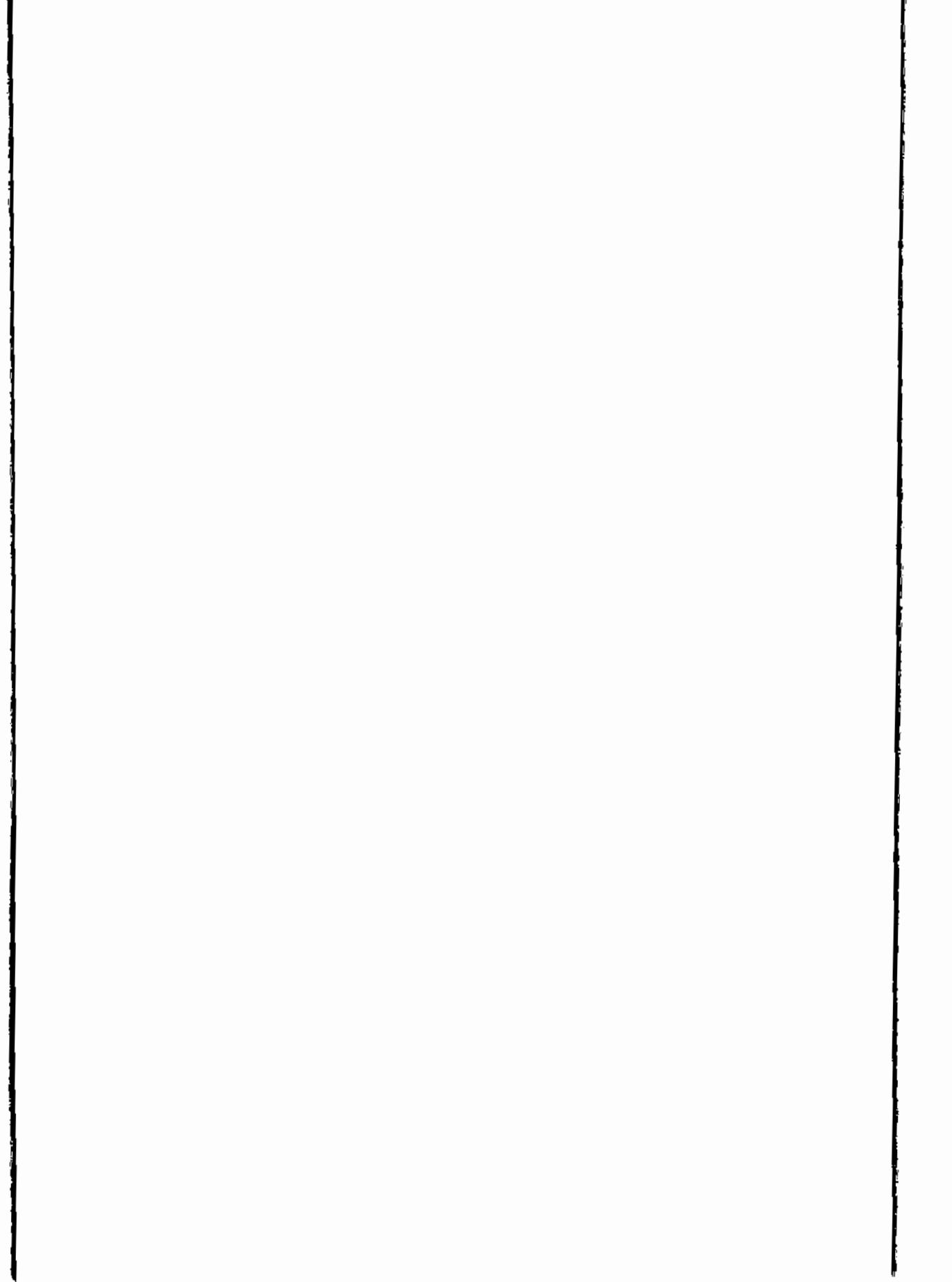




Fig. 4



Fig. 5

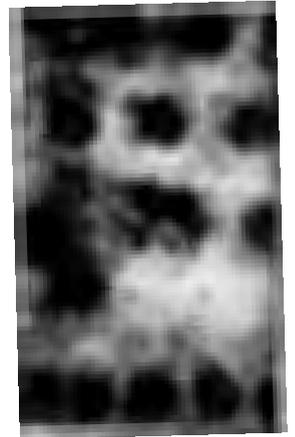


Fig. 6

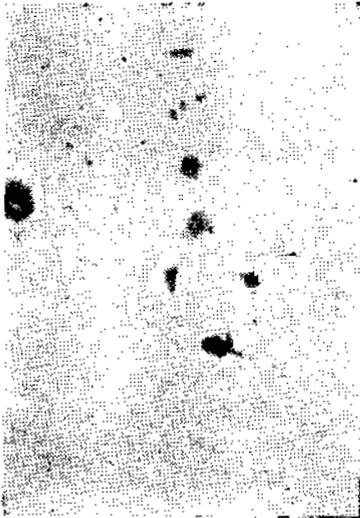


Fig. 7

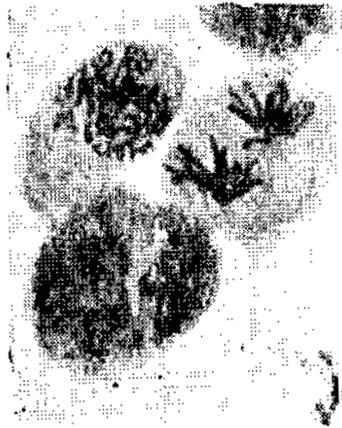


Fig. 8



Fig. 9

Figs. 4-9. Different anaphase types, Sticky bridges and lagging chromosomes (Figs. 4 & 5), complete disturbed anaphase (Fig. 6), tripolar anaphase (Fig. 7) loss of chromaticity (Figs. 8 & 9) resulting from treatments with 16 kr and 20 kr.