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**Cytoplasmic Changes Induced in the Walker Rat Carcinoma 256
by X-radiation of Different Intensities.**

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A comparison of the cytoplasmic changes induced in the Walker rat carcinoma 256 by different types and dosages of radiation revealed disorganization in the Golgi apparatus and little, if any, change in the mitochondria.¹ In that series of experiments it was noted that the enlargement of the Golgi region in the disorganization process seemed to bear some relationship to the dosage of irradiation, the greater the dosage the more marked the enlargement and density of the blackened mass. A subsequent series of experiments was carried out to test the effect of a variation in the milli-ampereage with all other factors unchanged as nearly as possible from the earlier series.

The disorganization of the Golgi apparatus and mitochondrial changes have been reported by several workers based on studies on the effects of radiation.²⁻⁵ Heilbrunn's⁶ review of the literature in Duggar's *Biological Effects of Radiation* shows that radiation has a destructive effect on mitochondria. It is interesting to note that Emmel⁷ noted a period of fragmentation of the Golgi apparatus and a darker staining osmic impregnated cytoplasmic area in the cells of the Ringer perfused frog's kidney.

In general the same procedure was followed as in the previous experiments. Rats bearing 10-day-old tumors were used for irradiation. Only the rats bearing uniformly growing, hard, non-necrotic tumors were used. The tumor growth characteristics are well known and came from the laboratory's continuously inbred strain of Sloniker white rats. No premature death of animals or

¹ Fogg, L. C., and Warren, S., *Am. J. Cancer*, 1937, **31**, 567.

² Guyer, M. F., and Claus, P. E., *Anat. Rec.*, 1934, **61**, 57.

³ Bartelmez, G. W., and Bensley, C. M., *Special Cytology*, edited by E. V. Cowdry, Paul B. Hoeber, New York, 1932, **3**, 1525.

⁴ Ludford, R. J., Tenth Scientific Report, Imperial Cancer Research Fund, 1932, p. 125.

⁵ Nahm, L. J., *J. Morph.*, 1933, **54**, 259.

⁶ Heilbrunn, L. V., and Mazia, D., *Biological Effects of Radiation*, edited by B. M. Duggar, McGraw-Hill, New York, 1936, **1**, 625.

⁷ Emmel, V. M., *Anat. Rec.*, 1938, **70**, 371.

change in the growth characteristics of the tumor, such as undue necrosis, occurred in this series of experiments.

The following is an outline of the experiments :

No. rats	Walker 256—10 day tumor.		Voltage
	Dosage		
6	800 r	at 15 MA	200 KV
6	800 r	at 20 MA	200 KV
6	800 r	at 25 MA	200 KV

For each series irradiated tumor bearing rats from each of the above series were killed at intervals of 2, 4, 6, 9, 12, 18 hours after onset of radiation. At each interval animals were killed by a blow to avoid the possible effect of anesthesia on tissue. As before the tumor was excised and small fragments of non-necrotic (*i. e.*, pearly white, somewhat hard and opaque) tissue were placed in fixative. Modified Mann-Kopsch method was used for revealing the Golgi apparatus and both Schridde and Regaud methods were used for the mitochondria. For this tissue these methods were adequate in that they revealed well stained cells.

A study of the effect of the radiation on the cytology of the cell confirms that of our earlier work.^{1, 8} In general, it causes a cessation of mitosis for a limited period, a variability in the ease of demonstration of mitochondria and a specific series of changes brought about in the Golgi apparatus. With a dosage of 800 r mitosis was completely retarded for a period less than 24 hours. The mitochondria underwent very little morphological change. The portion of the cell containing the Golgi net underwent the general sequence of changes, namely, the normal compact net, the spreading, the fragmentation, the spreading away from the original locus, the deeper staining cytoplasm and the gradual recovery into a compact net again. With a dosage of 800 r the massive dark abundance of osmic reducing substance is not so intense at the time when the cell is beginning to recover as it is when a dosage of 1600 r was given.

The cytology of the tumor cells in reference to the time of mitosis, the appearance of the Golgi and of the mitochondria can be noted in Table I.

It may be noted that there is little difference in the morphological changes that can be detected after this dosage at the 3 milliamperages listed, which represent those within the ordinary range of therapeutic radiation. Possibly the series at 25 MA shows the least effect or perhaps the quickest recovery.

There seems to be a period previous to the recurrence of mitosis in which the mitochondria have a greater susceptibility to stain and that

⁸ Warren, S., *Am. J. Roentgenol.*, 1937, **38**, 899.

TABLE I.

Hr after onset of radiation	15 MA	20 MA	25 MA
2 hr	Typical Golgi net. No mitoses. Mitochondria unchanged.	Typical Golgi net. No mitoses. Mitochondria unchanged.	Typical Golgi net. No mitoses. Mitochondria unchanged.
4 hr	Golgi net shattered. No mitoses. Mitochondria unchanged.	Fragmentary Golgi skeins. No mitoses. Mitochondria unchanged.	Slight change in Golgi net. No mitoses. Mitochondria unchanged but susceptible to stain.
6 hr	Fragmented Golgi covering a wide area. Densely staining area in cell. No mitoses. Mitochondria unchanged, little susceptibility to stain.	Wide, granular, densely staining area in cell but net reorganizing. No mitoses. Mitochondria unchanged, little susceptibility to stain.	Wide, granular, densely staining area in cell but net reorganizing. No mitoses. Mitochondria unchanged, some susceptibility to stain.
9 hr	A compact Golgi net in some cells. Dark staining cytoplasm. No mitoses. Mitochondria unchanged but very susceptible to stain.	A compact Golgi net in some cells. Dark staining cytoplasm. No mitoses. Mitochondria unchanged but very susceptible to stain.	A compact Golgi net in most of the cells. Dark staining cytoplasm. Rarely a mitosis. Mitochondria unchanged but very susceptible to stain and absent in Golgi area.
12 hr	Many cells appear normal. Prophases and metaphase apparent. Golgi net in host cells. Mitochondria unchanged.	Same as in 15 MA.	Same as in 15 and 20 MA.

this period, as compared with our earlier work,¹ varies with the dosage. This also holds true for the appearance of mitoses and for the series of changes in the Golgi net. The significance of this cytoplasmic disorganization and subsequent reorganization in relation to dosage remains for subsequent analysis. It is clear, however, that the Golgi in these experiments seems always to reorganize previous to any change in mitotic activity.

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