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Variation of Radiation Sensitivity of Paramecium aurelia as a Function of Time of Irradiation in the Interdivision Growth Cycle

THE larger ciliate protozoa provide a system which is particularly favourable for the investigation of the relationship between radiation sensitivity and events in vegetative growth and division. Individual animals can be isolated in the visible stages of cell division in sufficient numbers to provide a synchronized population for experimentation. Synchronization in this manner produces a minimum of the growth abnormalities which occur when synchronization is produced by such means as heat shock or antibiotics.

This type of synchronization has permitted the determination of the time of DNA synthesis¹ in Paramecium aurelia during interdivisional growth as well as the course of dry weight increase². Kimball and his associates have also shown that the sensitivity of P. aurelia to the mutagenic effects of ionizing radiation varies greatly during the interdivision growth cycle³, and that this variation approximately coincides1 with the beginning of DNA synthesis in the macronucleus which occurs about halfway through the interdivision period. The relationship of the lethal effects of X-rays and ultra-violet light to time of irradiation in the interdivision cycle can be determined in cultures synchronized in the manner already indicated.

A strain of *P. aurelia* which was isolated from Texas water was grown in lettuce media with Escherichia coli B/r as the food organism. Animals were selected during their cellular division and were allowed to grow in normal media to the desired phase for radiation. Interdivision times were approximately $6\frac{1}{2}$ h and were determined exactly for each experiment. The X-ray source was a General Electric 'Maxitron' (half-value layer 4.5 mm aluminium). The dose rate was determined by ferrous sulphate chemical dosimetry to be 15 krads/min. Ultraviolet light from General Electric germicidal lamps (2537 Å) was delivered at dose rates of $10-40 \text{ ergs/mm}^2/$ sec; these dose rates were determined by a calibrated photocell. Following irradiation, isolated animals were placed in depression slides and were observed until large cultures were produced or until the line died out

The maximum number of animals which could live in a depression slide was determined by the amount of food supplied; usually the number ranged from 40 to 200 animals. Sixteen progeny was the minimum number of progeny to be considered a survivor; however, after more than four progeny had descended from an irradiated individual they continued to grow until limited by the food supply.

The survival results were analysed to determine the LD_{50} and the 95 per cent confidence limits of LD_{50} using

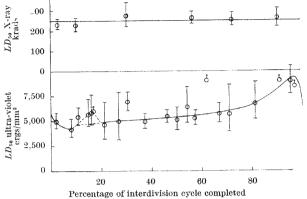


Fig. 1. Variation of LD_{29} Parameeium aurelia irradiated with X-rays and ultra-violet light during the interdivision growth cycle. The cycle is considered to start with the separation of the dividing animals. Circles with upward pointing arrows are plotted at the highest dose used in experiments where less than half the animals died at the highest doses used. The LD_{90} is evidently near or above this dose-level. Lines showing the trend of the response are fitted by eye

a logit type analysis. The actual computations were done on an IBM 1620 electronic computer. The results are illustrated in Fig. 1; LD_{50} is plotted as a function of the percentage of the interdivision growth cycle which the animals had completed at the time of irradiation. About 100 isolated animals were used to determine each point in the figure. The separation of the two daughters was taken as the start of the cycle.

One may conclude that the X-irradiated animals show only slight variation in radiation sensitivity through the interdivision cycle. Ultra-violet irradiated animals tend to be most sensitive shortly after division and gradually increase in their resistance to radiation. X-ray sensitivity does not seem to be related to DNA synthesis; however. a relationship between ultra-violet sensitivity and DNA synthesis may be possible. Results obtained from the ultra-violet experiments were more erratic than those obtained from the X-ray experiments. This difference might be a consequence of a more uniform deposition of radiant energy throughout the body of the X-irradiated animals. The results reported here tend to support the hypothesis⁴ that animals requiring extreme doses of radiation, particularly ionizing radiation, may be killed by cytoplasmic radiation damage rather than by damage to the cellular DNA.

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BIOLOGY

Adaptation of Horse Sickness Virus to Tissue Culture

IN a previous paper it was briefly stated that the horse sickness virus (HSV) may be adapted to the hamster kidney cells¹. The purpose of this communication is to describe the interaction between virus and cells and the development of infectivity during a single growth-cycle of the virus.