CONDITIONS FOR DISTANT INTERCELLULAR INTERACTION DURING ULTRAVIOLET RADIATION

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The objects of the investigation were to study the role of uv radiation in distant intercellular interactions (DII) and the conditions for obtaining a "mirror" cytopathic effect (MCPE). An extremal state of the cells in the radiating culture caused by uv-radiation was shown to induce a distant cytopathic effect (CPE) in an intact detector culture in optical contact only with it, reflecting the specific character of the morphological features recorded in the affected culture. Preliminary uv-irradiation of the detector cells facilitates manifestation of the MCPE.

KEY WORDS: distant intercellular interactions; "mirror" cytopathic effect; uv radiation; cell culture.

In 1966, Kaznacheev et al. showed that cells in tissue culture (detector culture), grown in special chambers on quartz supports, capture the distinguishing features of radiation from

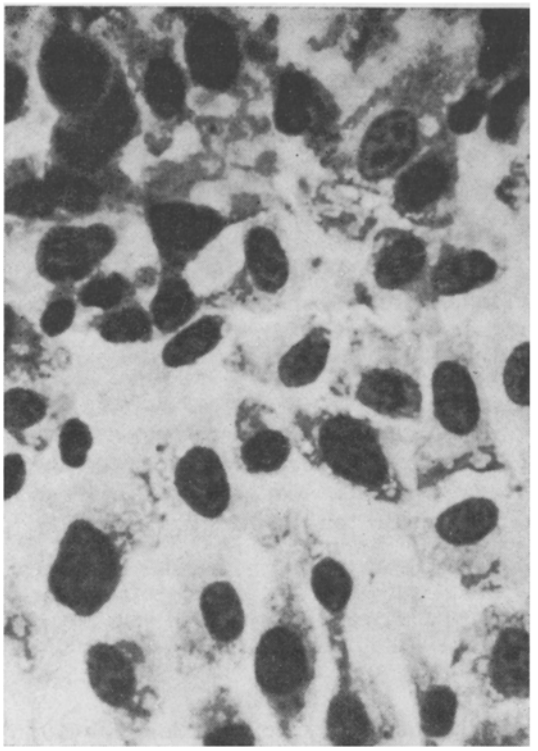

Fig. 1

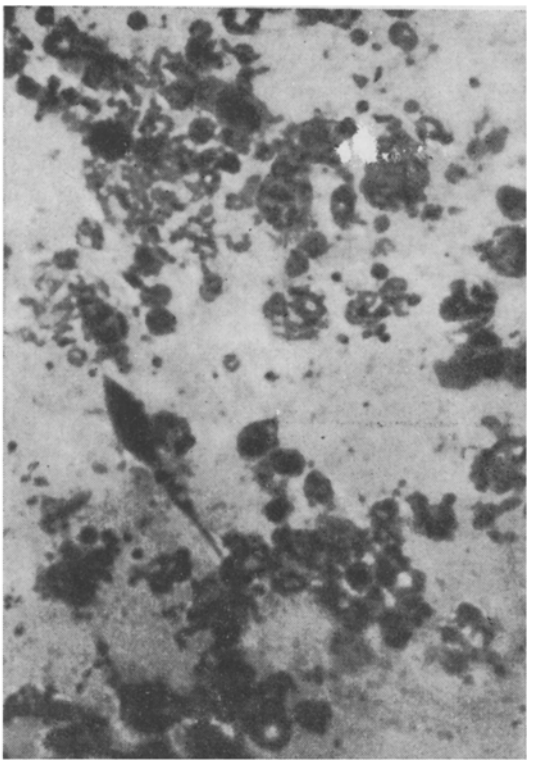

Fig. 2

Fig. 1. Control cell culture. Here and in Figs. 2 and 3: hematoxylin-eosin, 280×.

Fig. 2. Cell culture after uv irradiation.

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TABLE 1. Reproduction of MCPE in FECh Cell Culture on Contact with UV-Irradiated Culture

Experimental conditions	Number of experiments	Support	Duration of uv-irradiation, sec	Chamber A, CPE, uv	Chamber B, MCPE
Control	100	Quartz	—	—	—
	—	Simple glass	—	—	—
	100	"	45	100	—
Experiment	250	Quartz	45	250	191

Legend. Here and in Tables 2 and 3, chamber A is the inducer, chamber B the detector.

TABLE 2. Reproduction of MCPE in Hep-2 Cell Culture on Contact with UV-Irradiated Culture

Experimental conditions	Number of experiments	Support	Duration of uv-irradiation, sec	Chamber A, CPE, uv	Chamber B, MCPE
Control	100	Quartz	—	—	—
	—	Simple glass	—	—	—
	100	"	45	100	—
Experiment	250	Quartz	45	250	193

other cells (radiating culture), subjected to the action of extremal factors of biological and chemical nature (the "mirror" cytopathic effect — MCPE).

In this investigation the conditions for obtaining an MCPE under the influence of uv radiation on the radiating culture were studied.

Investigations were carried out in two directions: 1) reproduction of the CPE in a "mirror" detector cell culture during contact with a uv-irradiated radiating cell culture; 2) investigation of the MCPE in a detector culture previously irradiated with a small dose of uv radiation in distant intercellular interactions (DII) with a radiating culture infected with a virus.

EXPERIMENTAL METHOD

Distant intercellular interactions were studied in a system of cell cultures grown in isolated chambers so that only optical contact was preserved between the cell cultures through glass or quartz supports [1-4].

Cultures of Hep-2 cells (carcinoma of the human larnynx) and FÉCh cells (human embryonic fibroblasts) were used.

In the experiments of series I one of the cell cultures was irradiated with a lethal dose of uv radiation (40-45"), and an unirradiated cell culture served as the detector (the "mirror" culture). A BUV-30 lamp was the source of uv radiation. The dose of irradiation was determined experimentally for the actual lamp used.

In the experiments of series II the "mirror" culture was subjected to uv irradiation with a minimal dose (10-15") before contact with the radiating culture, infected with virus. Adenovirus of types 5, 23, and 25 was used. The minimal dose of irradiation (10-15"), not inducing morphological changes in the cell culture, was chosen experimentally.

After 2 days the chambers were disassembled, the glass supports with cells growing on them were sealed, and after fixation and staining, the cell cultures were examined morphologically.

The experiments were accompanied by an appropriate control with the aim of detecting spontaneous degeneration in the uninfected culture, by a control of the experimental conditions in which simple glass was used instead of the quartz support, and by passage of the virus from infected and uninfected chambers.

The CPE was judged from the ratio of the number of dying cells to the total number of cells and from the type of morphological changes.

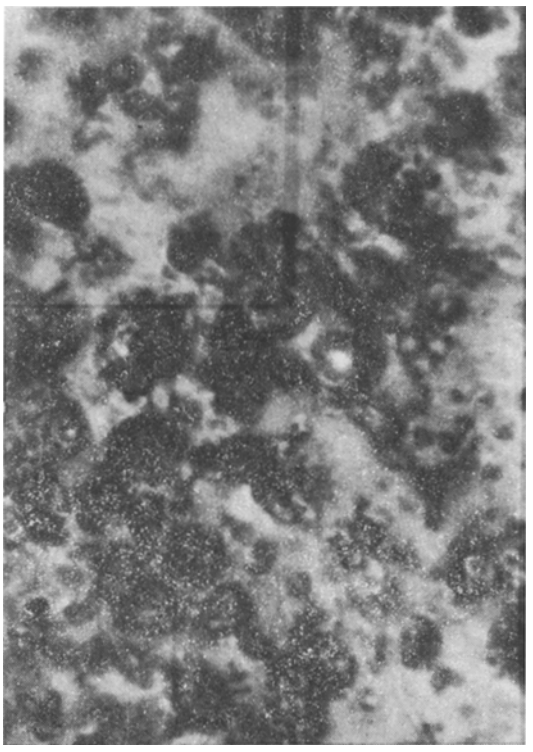

Fig. 3. "Mirror" cytopathic effect in cell culture.

TABLE 3. Reproduction of MCPE in FÉCh Culture by Means of Adenovirus After Preliminary Uv-Irradiation of "Mirror" Chamber

Type of experiment	Support	Duration of uv-irradiation, sec	Extremal agent (virus)	Chamber A, CPE (virus)	Chamber B, MCPE	Number of experiments
Control	Quartz	—	—	—	—	100
	Simple glass	—	—	—	—	100
UV dose control	Simple glass	15	—	—	—	100
	Quartz	15	—	—	—	100
Irradiation of "mirror" chamber B	Quartz	15	Adenovirus of types 5 and 23	159	158	159

EXPERIMENTAL RESULTS

Reproduction of the MCPE in the "mirror" detector culture on contact through the quartz support with the uv-irradiated radiating cell culture was observed in 384 of 500 cases (Tables 1 and 2, Figs. 1-3).

In the chambers in which simple glass was used as the support, no MCPE developed, whereas in the irradiated chamber a morphological picture characteristic of uv irradiation appeared. The CPE obtained by irradiation of the cell culture from the uv source corresponded to that described in the literature [5, 6]: Initially the cells lost their staining properties, the nuclei ceased to stain with basic dyes, and the nuclear material condensed into a dense hyperchromic body. The cytoplasm remained oxyphilic. The nuclear material then became pulverized and the cells were turned into round cytoplasmic formations, which later developed granular degeneration. In the case of a well-marked CPE the whole specimen consisted of an area littered with amorphous granular oxyphilic masses. In the "mirror" cultures the character of the morphological changes was identical. These results confirm the presence of distant intercellular interactions due to optical contact [1-4].

In the experiments of series II the role of small doses of uv radiation acting on the detector culture was studied during reproduction of MCPE by virus infection of the radiating cell.

The results of these experiments are given in Table 3. They show that irradiation of the cell monolayer in the "mirror" chamber with a minimal dose of uv radiation facilitated the appearance of MCPE.

According to data in the literature, if a stimulus acts on a cell population immediately after irradiation, its action is potentiated [7-12].

In the control experiments the cell culture irradiated in the same way remained undamaged. The results of the control tests with simple glass as the support were negative.

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CORRELATIONS BETWEEN MITOTIC INDICES OF BONE MARROW CELLS IN DOGS

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The mitotic indices of erythroblasts, basophilic and polychromatophilic normoblasts, myeloblasts, promyelocytes, and myelocytes were shown to decrease depending on the degree of differentiation of the cells in the ratio of 4:2:1. The values of the mitotic indices were shown to be an inverse power function of the number of cells.

KEY WORDS: mitotic indices; number of cells; correlation.

Many new methods by which the proliferation of bone marrow cells under normal and pathological conditions can be studied have recently been developed. In particular, the results of autoradiographic investigations and the results of investigation of cell proliferation in bone marrow cultures and in splenic colonies have been widely published [5, 9-12]. The results of detailed morphological investigations of bone marrow are sufficiently informative in this respect [4]. For instance, investigations of mitotic indices (MI) of whole bone marrow (per 1000 nucleated cells), of the myeloid and erythroid series (per 1000 erythronormoblasts and granulocytes), and in 100 erythroblasts, basophilic and polychromatophilic normoblasts, myeloblasts, etc., reflect the intensity of division of bone marrow cells.

Parallel determination of the ratio between different forms of cells of the erythroid and granulocytic series (erythrocyte and granulocyte counts) makes it possible to record activity of the influx of committed cells from the higher division.

The study of correlation between mitotic indices and the relative number of cells in erythrocyte and granulocyte counts can shed light on the existence and character of any significant correlation between these indices.

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