THE USE OF ULTRA-VIOLET FLUORESCENCE MICROSCOPY IN THE STUDY OF THE ANTIBLASTOMA PROPERTIES OF THE BLOOD SERUM

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The ability of blood serum from healthy individuals to lyse tumor cells has been described in a number of publications [5,6,8,9]. This property of the serum, commonly known as the cancerolytic ability, is absent in individuals suffering from malignant tumors. There have been apparently two reasons why the phenomenon of cancerolysis has not been made use of in clinical practice; these are a relative lack of precision in the method of observation of the cell lysis, and the fact that the cancerolytic properties of the blood are reduced in cancer patients to the same extent as in patients suffering from certain other conditions.

I. E. Brumberg and E. M. Brumberg [4] have recently described a new method of observing the damaging effect of blood on tumor cells; this method is based on the observation by the authors that the intensity of ultraviolet (UV) fluorescence of tumor cells is decreased when they are placed into the plasma or serum of healthy animals or humans. It was also determined that the plasma and blood serum from humans and animals suffering from malignant tumors cause less inhibition of the UV fluorescence of tumor cells than the plasma or serum of healthy humans and animals. This method proved to be more sensitive and gave more definite quantitative results than that based on the observation of cell lysis. Because UV fluorescence microscopy allows the observation of early stages of cell damage in the absence of their lysis, we are calling this property of the blood not cancerolytic ability, but by a more general term, the antiblastoma property of blood.

In the above-mentioned publication [4], the experiments were conducted mainly on white mice with only preliminary attempts to apply them to humans.

The present communication presents results of further experiments in this field, conducted on a large number of patients. So far we are able to express only preliminary considerations regarding the possibility and the nature of the utilization of this method in clinical practice.

METHODS

The UV fluorescence of tumor cells was observed with the aid of a special fluorescence microscope which transmits ultra-violet light. The apparatus and the methods of preparation of slides for UV fluorescence microscopy were described in our previous publications [2,3]. Ehrlich's ascites cells were used in these experiments. Blood to be tested was obtained by venipuncture from patients or donors. Tumor cells were placed for 30 min at room temperature into 0.5-1 ml of blood serum. Following this, the drop of blood serum together with the living tumor cells suspended in it, was placed on a quartz glass slide and covered with a quartz glass cover slip, and studied through the UV fluorescence microscope. A special photoelectric installation described previously [1] permitted the photometry of fluorescence of individual tumor cells, selected in the field of vision. The fluorescence was induced by long

wavelengths of 250-280 mμ and photometry was also done within the long wave range of 340-360 mμ. The most intense natural fluorescence of most animal cells occurs within this region of the spectrum. The fluorescence of cells was measured in relation to a piece of fluorescent glass whose intensity of fluorescence was taken as a unit.

Blood sera from 120 cancer patients and from 200 healthy donors were studied in these experiments.

RESULTS

Blood sera from healthy individuals lowered the intensity of UV fluorescence of Ehrlich's ascites cells placed in it by 40-45%, as compared with the fluorescence of control cells placed in normal saline under the same experimental conditions, following their removal from the abdominal cavity of mice.

Blood sera from cancer patients did not alter the intensity of fluorescence of tumor cells placed in it, or lowered the fluorescence of cells not more than 10-12% in some cases. The differences in the effects produced by blood sera of healthy donors and cancer patients exceeded considerably the limits of the precision of the method.

The intensity of UV fluorescence of Ehrlich's ascites tumor cells, following incubation for 30 min in normal saline, was equal to 0.36 ± 0.001 arbitrary units, in blood serum from cancer patients, 0.34 ± 0.001, and in blood serum from healthy donors, 0.19 ± 0.04 arbitrary units.

It is significant that, in none of the cancer patients studied, the intensity of fluorescence of ascites cells was lowered by more than 12%. Of interest also was the absence of differences in the effect of the blood serum from cases of different localizations of malignant tumors and of the clinical conditions of the patients.

The nature of antiblastoma action of the blood serum is not clear. We are possibly dealing with a mechanism which is related not only to the defense reaction of the organism against tumor growth, but with that which may be a regulatory factor of a more universal biological significance. While we are not attempting at this time to express a definite opinion on this problem, we are presenting certain recent experimental results having a direct bearing on it.

When too much ascitic fluid was added to the blood serum from healthy donors, there was less lowering of the fluorescence of the tumor cells. When the cells were washed in normal saline before they were added to the serum, the results obtained were practically independent of the number of the cells added. This indicates that tumor cells secrete into the surrounding medium a substance or substances which are able to bind the neutralizing substances responsible for the antiblastoma effect, possibly in accordance with the antigen-antibody reaction type.

In relation to the above, it must be noted that apparently only the antiblastoma blood factor has a direct effect on tumor cells. This deduction is based on the following: while blood plasma of healthy humans and animals lowers the intensity of UV fluorescence of Ehrlich's ascites cells placed in it, the ascitic fluid itself has no effect on the fluorescence of these cells and they fluoresce with the same intensity in this fluid as in normal saline. It seems that the substance, which we will call the blastoma factor, (to be differentiated from the antiblastoma blood factor), has no direct effect on tumor cells, and the part played by it in the above-mentioned experiments, as well as in the organism of the cancer patient, is reduced to chemical binding and deactivation of the antiblastoma factor, or to an inhibiting action on the mechanism which produces it.*

The results of the experiments conducted by us allow us to make some preliminary conclusions on the possibility of utilization of this phenomenon in oncology.

The normal antiblastoma action of blood serum which is expressed in the reduction of UV fluorescence of Ehrlich's ascites cells by not less than 30%, should apparently testify that a given individual is not suffering from malignant growths.

In order to accept or reject this method for making a positive diagnosis of malignant tumor processes, it is necessary to survey by means of this method a large number of individuals, in particular those suffering from different diseases, according to a definite program. At the present time, it is difficult to state how early in the course of the disease it is possible to detect the decrease of the antiblastoma action of blood serum by means of this method. Our experiments have shown that this effect is clearly defined already in the initial stage of cancer.

*Simultaneously with experiments in which we studied the effect of plasma or serum on the UV fluorescence of cells, we studied the inhibiting and stimulating action of spleen extract (antiblastoma factor) and of ascitic fluid (blastoma factor) on the development of tumors in mice. A parallelism between these phenomena was established. These experiments will be described separately in a later publication.

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All abbreviations of periodicals in the above bibliography are letter-by-letter transliterations of the abbreviations as given in the original Russian journal. *Some or all of this periodical literature may well be available in English translation.* A complete list of the cover-to-cover English translations appears at the back of this issue.
