ONCOLOGY

THE ANTIGENS OF THE CANCER CELL

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Research directed to the study of the antigens of malignant tumors has in the past occupied the attention of many oncologists. In work by L. A. Zilber and his co-workers [1], and also in research by P. N. Kosyakov [2], A. K. Saakov [3] and other writers, it was shown that in addition to specific cancer antigens the tumor contains a large number of proteins, similar or identical in their antigenic properties to normal tissue proteins.

In the present work we set out to investigate the presence of species, organ and cancer antigens directly in the cells of a malignant tumor, using for this purpose a suitable type of experimental cancer.

EXPERIMENTAL METHOD

The investigation was done on an ascitic variant of the Ehrlich mouse adenocarcinoma. Analysis of the antigenic properties of the tumor was performed by the anaphylaxis with desensitization method. In the experiments we used guinea pigs weighing 250-300 g. The guinea pigs were sensitized subcutaneously to the tumor by a dose of 15 mg. Desensitization and the determining injections of antigens in corresponding doses were performed intracardially 3-4 weeks after the first protein injection.

EXPERIMENTAL RESULTS

The use of an ascitic form of mouse cancer in our investigation had definite advantages since it permitted the analysis of antigens directly entering into the composition of the cancer cells. Malignant cells floating freely in the ascitic fluid could easily be freed from foreign protein by repeated washing with physiological saline, thus preventing it from taking part in the anaphylactic reactions. We assessed the degree of freedom from foreign protein of the cancer cells by the anaphylactogenic properties of the water used for washing. Preliminary experiments showed that washing the cells seven times produced maximum freedom from foreign proteins.

As may be seen from Table 1, guinea pigs sensitized by washing fluid in a volume of 0.05 ml*, showed no reaction on injection of 0.5-1 ml of original antigen.

The guinea pigs showed no reaction to the injection of 0.3 ml of mouse serum. However, subsequent injection into these guinea pigs of a saline extract of washed cancer cells produced clear anaphylactic shock in all the animals used in the experiment.

* A dose of 0.05 ml exceeds by 8 times the quantity of fluid present in one sensitizing dose of cancer cell precipitate.

TABLE 1

Antigenic Properties of the Supernatant Fluid obtained by Washing Ehrlich Mouse Adenocarcinoma Cells 7 Times

Serial No.	Guinea pig No.	Sensitization	Desensitization										
		7th wash adenocarc	7th wash adenocarc cells		Mouse serum						Mouse adenocarcinoma		
		Dose (ml)	Dose (ml)	Reaction	Dose (ml)	Reaction	Dose (ml)	Reaction	Dose (ml)	Reaction	Dose (mg)	Reaction
1	2985	0.05	0.5	—	0.3	—	·	·	·	·	50	++
2	3354	0,05	1.0	—	0.3	—	·	·	·	·	50	+++
3	2591	0.05	1.0	—	0.3	—	·	·	·	·	50	+
4	3853	1.00	1.0	—	0.3	++	0.3	++	0.3	—	50	+
5	3920	1.00	1.0	±	0.3	+	0.3	+	0.3	—	50	+++
6	3958	1.00	1.0	±	0.3	+	0.3	—	0.3	—	50	+++
7	3789	1.00	1.0	—	0.3	++	0.3	++	0,3	—	50	++

Conventional signs: — no reaction; + brief nose scratching with the paw; ++ stronger nose scratching, sneezing, shortness of breath, bristling of the fur, cough; +++ the same but more pronounced, passage of urine and feces; ++++ convulsions, leaping about, spasms, terminating usually in death of the animal; · no injection given.

A similar pattern could be seen in guinea pigs sensitized to washing fluid injected in a volume of 1 ml. In this case the guinea pigs remained insensitive to the original antigen but showed a feeble reaction to mouse serum and reacted to a greater degree to injection of tumor extract.

Thus the experimental results show that repeated washing of cells of ascitic cancer with physiological saline largely frees the tumor cells from foreign protein, in particular from proteins which are similar to the serum proteins in their antigenic structure, i. e. to the specific proteins of normal tissues. The experiments also showed that the washings contained a considerable quantity of protein of related origin to intracellular tumor proteins. We may suppose that during washing of the cancer cells there is a gradual removal of normal proteins and a parallel accumulation of tumor proteins which is possibly due to destruction of some of the tumor cells during washing.

The possibility of freeing tumor cells from foreign proteins and, consequently from risk of causing anaphylactic reactions, enabled us later on to make a series of experiments to analyze the antigenic properties of the proteins directly entering into the composition of malignant cells. We were particularly interested in the question of whether the proteins of the malignant cell possess any species antigenic specificity. For this purpose we sensitized a group of guinea pigs by subcutaneous injections of 15 mg doses of washed cancer cell precipitate. As antigen, to test the specific properties of the cancer cell protein, we used mouse serum.

The experiments showed (Table 2) that mouse serum, injected intracardially into the guinea pigs, caused an anaphylactic reaction in all the animals. A selected dose of 0.5 ml of serum proved adequate to desensitize the animals to serum protein on the first injection, since on repeating the injection of serum the guinea pigs showed no reaction. As we might have expected, desensitization of the guinea pigs to serum protein did not remove their sensitivity to subsequent injection of the original antigen. After injection of extract of cancer cells, 2 of the 4 animals used in the experiment died from anaphylaxis and the remaining two survived severe anaphylactic shock.

Guinea pigs in the second group, sensitized to mouse cancer cells but not desensitized to mouse serum, reacted violently to injection of cancer cell extract so that all the animals died as a result from anaphylactic shock. Unsensitized guinea pigs did not react to injection of these particular antigens (control of the toxicity of the antigen).

The results of this experiment show that proteins which in their antigenic properties are close to the proteins of mouse serum enter into the composition of the cancer cells. In other words, the intracellular protein of a mouse tumor possesses the species properties of the normal tissue proteins.

TABLE 2

Species Antigen in Ehrlich Adenocarcinoma Cells.

Serial No.	Guinea pig No.	Sensitization: Mouse adenocarc. Dose (mg)	Desensitization: Mouse serum Dose (ml)	Reaction	Dose (ml)	Reaction	Determining injection: Mouse adenocarc. Dose (mg)	Reaction
Experiment								
5	2700	15	0.5	++++	0.5	—	50	++++
6	1103	15	0.3	+++	0.5	±	50	++
7	1350	15	0.5	+++	0.5	—	50	Fatal shock
8	3333	15	0.5	+++	0.5	—	50	The same
Control of sensitization								
1	1793	15	.	.	.	.	50	" "
2	2865	15	.	.	.	.	50	" "
3	1232	15	.	.	.	.	50	" "
4	2116	15	.	.	.	.	50	" "
Control								
9	3032	.	0.5	—	0.5	—	50	—
10	1773	.	0.5	—	0.5	—	50	—
11	2119	.	0.5	—	0.5	—	50	—

Conventional signs the same as in Table 1.

The fact that desensitization of guinea pigs to species protein does not remove, but only to some extent weakens, their sensitivity to subsequent injection of the original protein is evidence of the antigenic complexity of the cancer cell.

Bearing in mind that the tumor serving as the object of our investigation is related in origin to the spontaneous breast cancer of mice, we may suppose that besides the species antigen, this tumor includes in its composition protein which, in its antigenic properties, is identical or close to the organ antigen of mouse breast cancer. In order to test this hypothesis we made experiments in which as a desensitizing agent we used mouse breast tissue.

As also in the previous experiment, the guinea pigs were sensitized to cells of the mouse tumor. Desensitization was first brought about by mouse serum and later by an extract of mouse breast. As a determining injection the guinea pigs received the original antigen. The experiments showed (Table 3) that guinea pigs, fully desensitized to serum protein, clearly reacted by anaphylactic shock to the injection of an extract of mouse breast. However, after full desensitization to this antigen also, they retained their sensitivity to the original antigen. On injection of extract of cancer cells, all the animals in the experimental group developed severe anaphylactic shock. Controls, in the form of unsensitized animals, showed no reaction whatever on injection of these antigens.

Analysis of the antigenic properties of the tumor by the use of breast tissue therefore showed that among antigens taking part in the composition of the cancer cell are some which are similar in their properties to breast antigen. It is possible that in the cancer cell may be found proteins which, in their antigenic properties, are similar to proteins of other organs and tissues of mice.

These facts are evidence that in a cancer cell are found proteins, similar or identical in their antigenic properties to proteins of mouse serum and breast. In addition, as the experiments showed, along with species

TABLE 3

The Presence of Organ Antigen in Ehrlich Adenocarcinoma Cells

Serial No.	Guinea pig No.	Sensitization: Mouse adenocarc. Dose (mg)	Desensitization: Mouse serum Dose (ml)	Reaction	Dose (ml)	Reaction	Dose (ml)	Reaction		Dose (ml)	Reaction	Dose (ml)	Reaction	Mouse breast Dose (mg)	Reaction	Dose (mg)	Reaction	Determining injection: Mouse adenocarc. Dose (mg)	Reaction
Experiment																			
1	2189	15	0.3	++++	0.5	±	0.5	—	Interval 20 hours	0.5	++++	0.5	++	30	+++	30	±	50	++++
2	2775	15	0.3	++++	0.5	+	0.5	—		0.5	—	.	.	30	++	30	±	50	++++
3	2200	15	0.3	++++	0.5	±	0.5	—		0.5	—	.	.	30	+++	30	±	50	++++
4	1989	15	0.3	+++	0.5	+	0.5	—		0.5	++	0.5	—	30	++	30	±	50	Fatal shock
Control																			
5	3071	.	0.3	—	0.5	—	0.5	—		0.5	—	0.5	—	30	—	30	±	50	—
6	3391	.	0.3	—	0.5	—	0.5	—		0.5	—	0.5	—	30	—	30	—	50	—
7	3118	.	0.3	—	0.5	—	0.5	—		0.5	—	0.5	—	30	—	30	—	50	—

Conventional signs the same as in Table 1.

and organ antigens the tumor contains a whole complex of other proteins, among which are those which are and those which are not found in normal organs and tissues, the latter being regarded as specific tumor proteins.

SUMMARY

Ascitic variation of mice adenocarcinoma served as an object of investigation. Analysis of antigenic properties of the tumor was conducted by the method of anaphylaxis with desensitization on guinea pigs. Repeated washing of floating cells of mice cancer from foreign proteins gave the opportunity to investigate the antigens which constitute the content of the tumor cells. The presence of proteins which are similar or identical to the proteins of the serum and the mammary glands was demonstrated in cancer cells. Besides species-specific and organ-specific antigens, numerous other antigens are found in cancer cells, among which the specific tumor antigen is evidently present.

LITERATURE CITED

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* In Russian.