porting measure during the pharmacotherapy, radiotherapy, or immunotherapy of tumors.

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HOMOLOGOUS ANTITISSUE ANTIBODIES AS A FACTOR INHIBITING THE DEVELOPMENT OF MALIGNANT TUMORS

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Experiments with transplanted and induced tumors showed that the physiological system of autoimmunity (homologous antitissue antibodies) can be used to increase resistance of the host to growth of malignant neoplasms. A method is developed for obtaining a globulin preparation containing normal antitissue antibodies in high titer.

KEY WORDS: *resistance; autoimmunity; normal antibodies; carcinogen.*

The attention of research workers is at present being increasingly drawn toward the use of natural immunological mechanisms of defense against the appearance and growth of malignant neoplasms. The use of normal antitissue antibodies, concerned in the regulation of metabolic processes, the removal of products of tissue metabolism and tissue breakdown, and also performing other no less important physiological functions, can be emphasized in this respect [1, 2, 6-8, 11-13].

During growth of a malignant neoplasm continuous autoimmunization of the host takes place under the influence of breakdown products arising from the tumor. In Klemparskaya's opinion, autoimmunization inhibits the response to the specific tumor antigen and may be one of the causes preventing rejection of the tumor.

The immunodepressive effect of tissue autoallergy is well known [6]. One way of suppressing autoallergy is by injecting ready-made preparations of normal antitissue antibodies.

The object of this investigation was to develop a new experimental method of increasing resistance of the host to growth of a malignant neoplasm by injection of special prepara-

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TABLE 1. Effect of Homologous Antitissue Antibodies on Growth of Ehrlich's Adenocarcinoma (50th day after transplantation)

Mice	Group of animals	Treatment of animals	Number of animals	Percentage developing tumors	Mean volume of tumors, cm^3 (M ± m)	% of regression of tumors	% of animals which died
Noninbred	1st control	Transplantation of Ehrlich's adenocarcinoma	20	100	3.88 ± 0.13	0	60
	2nd control	Transplantation of Ehrlich's adenocarcinoma + injection of globulin from intact mice	21	100	4.04 ± 0.21	0	61.9
	Experimental	Transplantation of Ehrlich's adenocarcinoma + injection of globulin from hemostimulated mice	20	85	0.35 ± 0.05	47	0
Strain C3HA (high predisposition to cancer)	1st control	Transplantation of Ehrlich's adenocarcinoma	21	100	4.38 ± 0.17	0	66.3
	2nd control	Transplantation of Ehrlich's adenocarcinoma + injection of globulin from intact mice	20	100	3.45 ± 0.13	0	55
	Experimental	Transplantation of Ehrlich's adenocarcinoma + injection of globulin from hemostimulated mice	20	90	0.10 ± 0.02	38.8	0
Strain C57BL (low predisposition to cancer)	1st control	Transplantation of Ehrlich's adenocarcinoma	19	90	2.29 ± 0.09	0	47.4
	2nd control	Transplantation of Ehrlich's adenocarcinoma + injection of globulin from intact mice	20	90	1.72 ± 0.07	0	40
	Experimental	Transplantation of Ehrlich's adenocarcinoma + injection of globulin from hemostimulated mice	20	85	0.45 ± 0.07	47	0

Legend. Differences between 1st and 2nd control groups of animals of the same strain were not significant ($P > 0.05$); differences between both control groups and experimental group of the same strain of animals were significant ($P < 0.001$).

TABLE 2. Effect of Homologous Antitissue Antibodies on Growth of Induced Tumors (150th day after injection of carcinogen)

Mice	Group of animals	Treatment of animals	Number of animals	Percentage developing tumors	Mean volume of tumors, cm^3 (M ± m)	% of regression of tumors	% of animals which died
Noninbred	1st control	Injection of MC	14	71.4	5.75 ± 0.8	0	40
	2nd control	Injection of MC + injection of globulin from intact mice	15	66.6	4.66 ± 0.7	0	30
	Experimental	Injection of MC + injection of globulin from hemostimulated mice	13	38.4	0.2 ± 0	60	0
Strain C3HA (high predisposition to cancer)	1st control	Injection of MC	19	68.4	4.99 ± 0.6	0	69.2
	2nd control	Injection of MC + injection of globulin from intact mice	19	68.4	3.81 ± 0.4	0	45.3
	Experimental	Injection of MC + injection of globulin from hemostimulated mice	18	38.8	0.30 ± 0.02	42.8	0
Strain C57BL (low predisposition to cancer)	1st control	Injection of MC	16	50	1.55 ± 0.3	0	37.5
	2nd control	Injection of MC + injection of globulin from intact mice	14	42.8	1.10 ± 0.1	0	33.3
	Experimental	Injection of MC + injection of globulin from hemostimulated mice	14	28.6	0.31 ± 0.03	25	0

Legend. Differences between 1st and 2nd control groups of animals of the same strain were not significant ($P > 0.05$); differences between both control groups and experimental group of the same strain of animals were significant ($P < 0.001$). MC) 20-methylcholanthrene.

tions of homologous antitissue antibodies. The foundations for this research were laid by the results of an investigation by Saraeva and Tereshchenko [9], who found that differences between strains in resistance to tumor growth depend on the initial level of normal antitissue antibodies, and who used with success a globulin containing normal antibodies to combat radiation autoallergy [10].

EXPERIMENTAL METHOD

Experiments were carried out on 264 sexually mature male mice of two strains: C3HA, with a predisposition toward the development of spontaneous and induced tumors, and C57BL/6, resistant to tumor development, and also on noninbred animals.

Transplantable Ehrlich's adenocarcinoma and a sarcoma induced by 20-methylcholanthrene (0.125 mg of the carcinogen in 0.2 ml olive oil) were used as the model with which to study antitumor activity of homologous antitissue antibodies. The tumor cells were transplanted and the carcinogen injected subcutaneously into the thigh.

To obtain the preparation containing normal antitissue antibodies in high titer, their formation had to be stimulated *in vivo* by procedures which did not introduce a heterologous antigen. Several such methods are known: injection of homogenates of homologous tissues, or of freshly obtained citrated blood from animals of the same species, and so on.

The method developed by Shal'nova was used for this purpose, namely giving one or two injections of homologous citrated blood in a dose of 0.25 ml. The animals were exsanguinated 3 days after the second hemostimulation and the serum obtained. The sera were titrated for antitissue antibodies by Hoigne's method in the modification of Klemparskaya and Raeva. Sera giving a reaction of 3 or 4 points were regarded as suitable.

Special investigations showed an increase in the titer of homologous antibodies in the blood of the hemostimulated animals as early as after 15 min, rising to a maximum after 3 days, and falling to the initial level again on the 8th day. The reaction to the second injection of blood continued to exhibit basically the same pattern while antibodies were being produced, but their circulation was a little prolonged. It should be noted that the second injection of homologous blood did not cause an increase in the antibody level compared with the first injection, as is the case during immunization with heterologous protein [4, 5].

The globulin fraction was separated from the pooled sera by the usual method, by precipitating with ammonium sulfate. The liquid globulin fraction, containing 20 mg protein in 1 ml, was frozen to −8 or −10°C and kept in the refrigerator at that temperature. The preparation was thawed at room temperature when required and used in the necessary dose.

The doses and intervals of injection of the preparation containing normal antitissue antibodies in high titer differed in the work with transplanted and induced tumors. In the work with transplanted tumors the most effective method was to inject globulin containing normal antitissue antibodies in a sessional dose of 0.05 ml (1 mg protein) twice: the first time, simultaneously with inoculation of the tumor cells, the second time, 10 days later.

In the work with induced tumors five injections of globulin from the hemostimulated animals were given, each dose consisting of 0.1 ml of the preparation (2 mg protein): the first injection was given simultaneously with the carcinogen, the rest at intervals of 30 days.

To confirm the connection between the antitumor effect and the presence of antitissue antibodies, a second preparation was obtained from the blood serum of intact animals (without hemostimulation). Hardly any normal antitissue antibodies could be detected in this preparation by Hoigne's method.

The time of appearance of the tumors, the dynamics of their growth or regression, and death of the animals were recorded.

Observations were made on the animals with transplanted tumors for 50 days and on animals with induced tumors for 5 months.

EXPERIMENTAL RESULTS

The effect of an increase in the resistance of the host to tumor growth is shown in Tables 1 and 2. Clearly injection of globulin containing normal antitissue antibodies in high titer caused some decrease in the percentage of development of tumors, a sharp reduction

in the size of those neoplasms which did develop compared with those in the control groups, and even complete regression of the tumors, which was never observed in the mice of the control groups, irrespective of the strain of the animals or the type of tumor (transplanted or induced) studied.

Among animals receiving injections of the preparation with normal antitissue antibodies, none died within the periods of observation specified above, whereas in the control groups the mortality among the animals varied from 30 to 69.2% depending on the strain of mice.

The experiments thus showed that injection of measured doses of homologous antitissue antibodies is an effective method of increasing resistance of the host to tumor growth.

It was not by chance that measured doses of normal antitissue antibodies were injected rather than activating the synthesis of normal globulins directly in the animal with the tumor. The reason for this was that the autoimmune response of the host to stimulation could differ in intensity depending on the initial state of immunological reactivity. Sudden activation of the immune response during tumor growth is just as undesirable (the phenomenon of immunological potentiation) as immunodepression.

The preliminary investigation showed that the antitumor effectiveness of homologous antitissue antibodies depends on the dose and rhythm of injection.

Consequently, in order to study the antitumor action of the system of normal antitissue antibodies, optimal doses and rhythms of their administration must be found.

The results of this investigation raise the question of the need for a further study of the role of homologous antibodies in the pathogenetic treatment of malignant neoplasms.

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