

A NEW APPROACH TO CANCER THERAPY*

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ABSTRACT

A new approach to cancer therapy is presented. It is based on the expected permeability of cancer cells to antibodies and ability of nucleic acid reactive antibodies to inhibit protein synthesis. The feasibility of the approach is shown with Yoshida ascites sarcoma in rats. When Yoshida ascites sarcoma was transplanted in rats preimmunized with bovine serum albumin conjugates of adenosine and uridine, a significant number of immunized rats survived after all the non-immunized rats had died. Tumor regression could be noted in a significant number of survivors.

INTRODUCTION

NUCLEIC acid reactive antibodies have been elicited and studied in several laboratories¹⁻⁴. Most of them bind to single stranded or denatured nucleic acids (DNA or RNA). These antibodies do not produce any ill-effect on normal cells in culture⁵⁻⁷, experimental animals⁸ or human beings⁹. There is overwhelming evidence in literature to the effect that normal mammalian cells are not permeable to antibodies¹⁰⁻¹³. All the transformed cells studied in culture have been found permeable to antibodies^{10,13}. So it is reasonable to expect cancer cells *in vivo* to be permeable to antibodies. If nucleic acid reactive antibodies which strongly bind to single stranded nucleic acids (for example, *m*-RNAs, *r*-RNAs and *t*-RNAs) get inside mammalian cells in sufficient numbers, they are potentially capable of inhibiting protein synthesis by making these nucleic acids non-functional. This can result in the death of these cells. Because of the permeability difference, these antibodies are likely to be detrimental to the cancer cells only and not to the normal cells.

As a test for the feasibility of the above approach, rats were immunized with protein-nucleoside conjugates and Yoshida ascites tumor was transplanted in them. A significant number of the immunized rats survived after all the non-immunized rats had died. Also tumor regression could be noticed in a few of the survivors.

MATERIALS AND METHODS

Bovine serum albumin and the ribonucleosides were from Sigma Chemical Co., U.S.A. Inbred Wistar male rats were from the Central Animal Facility of the Indian Institute of Science. Yoshida ascites sarcoma bearing rats were from the Tata Memorial Cancer Research Institute, Parel, Bombay.

Yoshida ascites sarcoma cells were maintained by intraperitoneal injection in inbred Wistar rats at 5 days intervals. About 3×10^7 cells/rat were used.

Bovine serum albumin (BSA) conjugates of adenosine (A) and uridine (U) were prepared by the method of Erlanger and Beiser¹⁴.

Effect of pre-immunization with nucleoside-protein conjugates on Yoshida ascites tumor in rats:

Inbred Wistar rats weighing about 80 gm were used. One group of rats were immunized with BSA-A and another group with a mixture of BSA-A and BSA-U. For this 200 μ g of BSA-A or a mixture of 100 μ g of BSA-A and 100 μ g of BSA-U in 0.1 ml saline was emulsified with 0.1 ml of complete Freund's adjuvant (CFA) and 0.1 ml injected i.p. and 0.1 ml s.c. in each rat. Simultaneous injection through two routes were given as it was expected to be more effective than single route injection. One set of controls was untreated whereas another set was given injections of CFA. The injections were repeated twice at 5 days interval. Yoshida ascites tumor (3×10^7 cells) were transplanted in all rats 5 days after the last injection.

RESULTS AND DISCUSSION

The results of the experiment are given in Table I. All the rats took the ascites tumor. Those rats which were not given any treatment before tumor transplantation died by the eighteenth day, whereas a significant number of rats in all the other three groups did not die indicating the good effects of the treatments given. The survivors were maximum (50%) in the group immunized with a mixture of BSA-A and BSA-U. The antitumor effect noted is attributed to the nucleic acid reactive antibodies elicited by the immunizations. It has been shown that when protein conjugates of adenosine or uridine are used for immunization of rabbits¹⁵⁻¹⁷ or rats (authors' observations), nucleic acid reactive antibodies are elicited. The effectiveness of the treatments would have depended on the concentrations of the nucleic acid reactive antibodies that would have been present in the animals during the period following tumor transplantation.

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TABLE I

Effect of immunization with protein conjugates of nucleosides on Yoshida ascites sarcoma in rats

	Treatment			
	Nil	CFA	BSA-A +CFA	BSA-A +BSA- U+CFA
Number of animals used	10	10	9	10
Number of "takes" of ascites tumor	10 (100)	10 (100)	9 (100)	10 (100)
Number alive on 19th day	0 (0)	3 (30)	4 (44)	5 (50)
Number alive on 35th day	0 (0)	2 (20)	1 (11)	5 (50)
Number developed solid tumor by 35th day	0 (0)	2 (20)	0 (0)	2 (20)
Number tumor regressed by 35th day	0 (0)	0 (0)	1 (11)	2 (20)*

Note: The numbers in the parenthesis indicate percentage.

*The fifth rat that was alive on the 35th day showed incomplete regression of tumor.

The tumor transplantation was done 5 days after the last immunization with the expectation that the nucleic acid reactive antibody titres would be the highest in the rats during the two weeks following tumor transplantation. The higher effectiveness of immunization with a mixture of BSA-A and BSA-U is expected to be due to the higher titres of nucleic acid reactive antibodies it is able to elicit compared to the adenosine conjugate alone. It seems reasonable to ascribe the noticeable effect shown by CFA to the elicitation of nucleic acid reactive antibodies by the killed mycobacterium cells in it. It is known that killed whole bacteria do produce nucleic acid reactive antibodies^{18,19}. It is also possible that one of the mechanisms by which BCG exhibits antitumor activity is through nucleic acid reactive antibodies.

Our preliminary studies⁴ using BSA conjugates of adenylic acid and adenosine showed that rats immunized with the adenosine conjugate rejected Yoshida ascites tumor to a significant extent when the tumor cells injected were less (10^7 cells) than that used in the present study (3×10^7 cells).

With the selection of better hapten carriers and the standardization of the dose, timing and route of immunization so as to get maximum elicitation of

nucleic acid binding antibodies, it is expected that it will be possible to improve the antitumor response in experimental animals. Anti-nucleic acid antisera could find use in passive immunization against cancer. The cancer therapeutic approach reported in this paper is expected to be applicable to all types of cancers permeable to antibodies in all vertebrates including man.

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