FUNCTIONAL ACTIVITY OF T AND B CELLS OF THE SPLEEN DURING TUMOR GROWTH

A. Sh. Norimov, R. M. Khaitov, and S. S. Gambarov

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The functional activity of B and T cells was investigated during tumor growth in mice. During tumor growth the ability of the B and T cells to interact on immunization with sheep's red cells was shown to be disturbed. B cells are affected sooner than T cells.

KEY WORDS: B cells; T cells; tumor growth.

As a rule, immunodepression develops in patients with malignant neoplasms and in experimental animals with tumors [4, 7]. It has been suggested that this phenomenon is due to the systemic action of the tumor on the organism [2].

In order to develop effective immune reactions against most (thymus-dependent) antigens interaction between the precursors of antibody-forming cells (B cells), derived from the bone marrow, and thymus-dependent (T cells) lymphocytes is necessary [8, 9]. It was shown previously that during tumor growth the immune response to the action of heterologous erythrocytes is depressed [3, 5].

In this investigation the ability of T and B lymphocytes to cooperate during immunization with sheep's red cells was studied in animals with developing tumors.

EXPERIMENTAL METHOD

Experiments were carried out on C57BL/6 mice into which cells of carcinoma Ca-755 were transplanted from syngenetic donors.

To study the effect of cooperation of T and B cells during the development of the immune response, 4-6 h after lethal irradiation (900 R) the recipient mice received an intravenous injection of bone marrow cells in a dose of $1 \cdot 10^7$ (B cells) and lymph node cells in a dose of $1 \cdot 10^6$ (T cells), obtained from tumor-bearing and intact mice, in different combinations. Bone marrow is known to contain only B cells, whereas the cell population of the lymph nodes consists mainly (65-70%) of T cells [8].

In some experiments the functional activity of T and B cells was studied in the spleens of tumor-bearing mice. For this purpose, spleen cells ($1 \cdot 10^7$) from tumor-bearing mice were injected either alone or together with bone marrow cells (to study the functional activity of the splenic T cells) or with thymus cells of intact mice (to study the functional activity of the splenic B cells) into lethally irradiated recipients. In these experiments bone marrow cells ($1 \cdot 10^7$) and thymus cells ($4 \cdot 10^7$) were added in sufficient number so that, when injected together, the same number of antibody-forming cells (AFC) was produced in the recipients' spleens as in response to the injection of $1 \cdot 10^7$ spleen cells from intact mice. Sheep's red cells, injected in a dose of $2 \cdot 10^8$ per mouse, were used as the antigen. On the eighth day after transplantation the number of AFC in the recipients' spleens was counted by Jerne's method [6]. The mice were irradiated in an absolutely lethal dose of 900 R on the "Stebel'-3A" apparatus.

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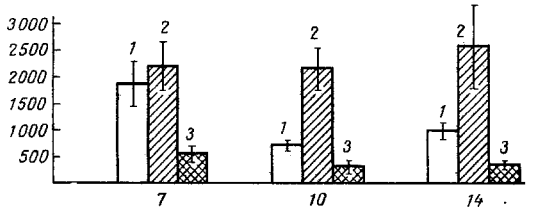

Fig. 1. Ability of B and T lymphocytes of tumor-bearing mice to interact cooperatively during immunization with sheep's red cells at different times after inoculation of carcinoma Ca-755. Ordinate, number of AFC in spleen; abscissa, time after inoculation of tumor (in days). 1) Intact bone marrow + lymph-node cells from tumor-bearing mice; 2) intact bone marrow + intact lymphocytes; 3) bone marrow from tumor-bearing mice + intact lymphocytes. Short vertical lines indicate 95% ($P \leq 0.05$) confidence limits.

The results were subjected to statistical analysis with calculation of the standard error of the arithmetic mean and the confidence interval I_p with a probability of 95% ($P \leq 0.05$).

EXPERIMENTAL RESULTS

As Fig. 1 shows, after combined injection of bone marrow (B cells) of intact mice and T cells obtained from tumor-bearing mice 7 days after inoculation with the tumor, approximately the same number of AFC was counted in the spleens of the syngenetic irradiated C57BL/6 recipients as after transplantation of B and T cells from intact animals.

However, after transplantation of intact B cells with T cells obtained from donors 10 days after inoculation with tumor cells, the number of AFC formed in the recipients' spleens was 67-71% less than the number of AFC counted after injection of B and T cells from intact mice.

After injection of a mixture of B cells from intact mice and T cells from mice bearing a tumor for 14 days, the number of AFC accumulating in the recipients' spleens was 60-67% less than after transplantation of B and T cells from intact donors.

Investigation of the functional activity of the B cells showed that they are damaged earlier than the T cells during induction of the tumor process. For instance, 7 days after transplantation of the Ca-755 tumor the B cells of these mice cooperated much less readily with intact T cells (Fig. 1). B cells obtained at later stages (10 and 14 days) after inoculation of the tumor were inhibited by a rather greater degree. However, the possibility could not be ruled out that the decrease in cooperative ability of the bone marrow and lymph node cells of tumor-bearing mice was due to the more intensive migration of B cells (from the bone marrow) and T cells (from the lymph nodes) into the spleen, for intensified migration of hematopoietic stem cells from the bone marrow is observed during tumor growth [1]. For this purpose, the functional activity of the T and B cells of the spleen of the tumor-bearing mice was studied.

As Table 1 shows, transplantation of spleen cells in a dose of $1 \cdot 10^7$ from tumor-bearing mice on the 7th and 10th days after inoculation of the tumor cells led to the formation of only one third as many AFC in the recipients' spleens as after injection of spleen cells from intact mice. On the addition of intact bone marrow ($1 \cdot 10^7$) to the spleen cells from mice bearing tumors for 7 days, approximately the same number of AFCs accumulated in the recipients' spleens as after transplantation of spleen cells from intact mice. Meanwhile, the addition of bone marrow cells to spleen cells taken from tumor-bearing mice on the 10th day after inoculation of the tumor did not cause any significant increase in the number of AFC in the spleens of the lethally irradiated recipients.

The absence of effect of addition of bone marrow cells to spleen cells of mice 10 days after inoculation with tumor cells was evidently connected with disturbance of the ability of the splenic T cells to interact with B cells during the development of the immune response to sheep's red cells.

Addition of thymus cells to spleen cells obtained from tumor-bearing mice on the 7th and also on the 10th day after inoculation of the tumor had no significant effect.

TABLE 1. Number of AFC in Spleens of Lethally Irradiated Recipients after Transplantation of Spleen Cells from Tumor-Bearing C57BL/6 Mice Alone or Together with Bone Marrow or Thymus Cells

Number of cells injected (in millions)				Number of recipients	Number M ± I_p ($P \leq 0.05$)
intact spleen	spleens of tumor-bearing mice	intact bone marrow	intact thymus		
10	—	—	—	12	2062±763
—	10*	—	—	6	670±320
—	10†	—	—	5	600±71,6
—	—	10	—	7	132±35,1
—	—	—	40	7	137,8±45,3
—	—	10	40	15	1882±334
—	10*	10	—	8	2003±845
—	10*	—	40	6	722,5±154,4
—	10†	10	—	7	858±582
—	10†	—	40		720±428

*Spleen cells obtained 7 days after inoculation with Ca-755.
†Spleen cells obtained 10 days after inoculation with Ca-755.

The results described above thus indicate that damage to B and T cells participating in the development of the humoral immune reactions takes place during tumor growth, and the B cells are damaged before the T cells.

It can accordingly be concluded that a functional change in the activity of the T and B cells participating in the development of humoral immune reactions may be one of the factors in the immunodepression developing in tumor-bearing animals. Such a disturbance of humoral immunity is evidently a reflection of the systemic action of the tumor on the host.

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