B SUPPRESSOR CELLS OF LYMPH NODES

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It is well known that the immune response is controlled by subpopulations of T cells [6, 10]. Meanwhile, it has been shown recently that cells possessing the characteristics of B lymphocytes can also exhibit immunoregulatory properties [1, 2]. The object of this investigation was to study the nature of suppressor cells in lymph nodes of intact mice.

EXPERIMENTAL METHOD

Hybrid (CBA × C57BL/6)F_1 mice from the "Stolbovaya" nursery, Academy of Medical Sciences of the USSR, were used.

Syngeneic bone marrow and thymus or lymph node cells, together with sheep's red blood cells ($2 \cdot 10^8$), were transplanted into lethally irradiated (900 R) recipients. Instead of bone marrow, some groups of mice received spleen cells from thymectomized, lethally irradiated mice, revived with bone marrow ($20 \cdot 10^6$ cells; B mice). In other experiments lymph node cells, before transplantation, were passed through a column with nylon gauze or were treated with anti-MBLA serum and complement or with anti-IgG serum and complement, or the mice donating the lymph nodes were treated with cyclophosphamide (200 mg/kg) 24 h before the cells were taken.*

On the 8th day after transplantation of the cells the number of antibody-forming cells (AFC) in the recipients' spleen was determined by Jerne's method [8]. The numerical results were subjected to statistical analysis with determination of the geometric mean and the upper and lower limits of its confidence interval at $P = 0.01$.

EXPERIMENTAL RESULTS

As Table 1 shows, between 6 and 7 times more AFC were formed in the spleen of the recipients after transplantation of spleen cells of B mice (B_S cells) and thymus cells than after transplantation of thymus and bone marrow cells. This was connected with the fact that, besides B cells, bone marrow also contains their precursors, which undergo maturation in the primary irradiated recipients (B mice).

A different picture was observed when lymph nodes were used as the source of T cells. After injection of B_S cells with lymph node cells, several times fewer AFC accumulated in the recipients' spleen than after transplantation of bone marrow and lymph node cells.

In the next experiments the lymph node cells were passed through a column with nylon gauze, which selectively retains B cells and allows T helpers to pass through.

Between 10 and 15 times more AFC were formed in the spleen of the irradiated recipients after injection of lymph node cells which passed through the column together with spleen cells of B mice than after transplantation of B_S cells together with unfractionated lymph node cells (Table 2).

When lymph node cells pass through the column, evidently the cells which suppress development of the cooperative immune response are retained on it.

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TABLE 1. Interaction between T and B Cells from Different Sources during Immune Response

Number of cells injected, millions				Number of recipients	Number of AFC
bone marrow	spleen of B mice	thymus	lymph nodes		
10	—	20	—	12	1314 (1073÷1611)
—	10	20	—	18	6702 (5653÷7945)
10	—	—	2	11	2705 (2042÷3579)
—	10	—	2	12	264,8 (194,5÷360,2)

TABLE 2. Ability of Lymph Node Cells Passed through a Nylon Column or Obtained from Donors Treated with Cyclophosphamide to Interact with Spleen Cells of B Mice

Number of cells injected, millions			Number of recipients	Number of AFC
bone marrow	spleen of B mice	lymph nodes		
10	—	2	12	2069 (1704÷2514)
10	—	2*	6	2391 (1681÷3401)
—	10	2	11	334 (261,3÷427,1)
—	10	2*	14	4575 (3906÷5338)
—	10	2†	12	2873 (2300÷3581)

*Lymph node cells were passed through nylon gauze columns.
†Donors of lymph node cells were treated with cyclophosphamide.

TABLE 3. Interaction of Lymph Node Cells Previously Treated with Anti-IgG or Anti-MBLA Serum and Complement with Spleen Cells of B Mice

Number of cells injected millions		Number of recipients	Number of AFC
spleen of B mice	lymph nodes		
10	2	14	355,3 (284,2÷444,2)
10	2*	11	1505 (1240÷1802)
10	2†	6	1834 (1219÷2761)

*Lymph node cells treated with anti-IgG serum and complement before injection.
†Lymph node cells treated with anti-MBLA serum and complement.

During transplantation of lymph node cells which passed through the column, together with bone marrow cells, the same number of AFC was formed in the recipients' spleen as after injection of intact lymph node and bone marrow cells. These results indicate that suppressors are not included in the system of adoptive transfer of bone marrow and lymph node cells.

As will be clear from Table 2, between 5 and 6 times more AFC accumulate in the recipients' spleen after injection of B_S cells together with lymph node cells from donors treated with cyclophosphamide than after transplantation of B_S cells with lymph node cells from intact mice.

Suppressor cells of lymph nodes thus adhere to nylon gauze and are sensitive to cyclophosphamide. These properties are those of B cells [5, 9] and also of the T suppressor subclass of T cells [4, 7].

TABLE 4. Effect of Dose of Bone Marrow Cells on Number of Antibody-Forming Cells Detectable in Recipients' Spleen

Number of cells injected, millions		Number of recipients	Number of AFC
bone marrow	lymph node		
10	2	6	1972 (2982÷1302)
20	2	7	3031 (4377÷2099)
40	2	8	5417 (7049÷4162)
80	2	6	820,8 (992,8÷678,9)

Treatment of the lymph node cells both with anti-MBLA and anti-IgG serum and with complement before their transplantation with B_S cells led to a four- to fivefold increase in the number of AFC accumulating in the recipients' spleen (Table 3).

The results are evidence that the suppressor cells of lymph nodes carry determinants characteristic of B cells on their surface. It must be pointed out that the possibility of expression of antigens of B cells on T suppressor cells was demonstrated previously [3]. However, as the present results show, suppressor cells of lymph nodes do not simply possess individual properties of B cells, which may also be present in T suppressors, but the whole assortment of their properties.

Inclusion of lymph node suppressor cells in the system of adoptive transfer takes place when the spleen, but not bone marrow, of B mice is used as the source of B cells. The appearance of the signal for inclusion of the suppressors may be due either to a higher concentration of B cells in the spleen of the B mice or to a difference in their quality (compared with bone marrow).

If the sending of the signal by the B cells was connected with their concentration, an increase in the dose of bone marrow in the adoptive transfer system of bone marrow and lymph node cells ought to lead to activation of the lymph node suppressor cells.

As Table 4 shows, an increase in the number of bone marrow cells within certain limits ($10 \cdot 10^6$ to $40 \cdot 10^6$) led to an increase in the number of AFC in the recipients' spleen. However, a further increase in the dose of bone marrow cells ($80 \cdot 10^6$) led to a marked decrease in the number of AFC.

The results of these investigations thus show that lymph nodes of intact mice contain suppressor cells switched on by a signal given out by B cells. These suppressor cells have the properties of B cells: They adhere to nylon gauze, they are sensitive to cyclophosphamide, and they carry antigenic determinants of B cells (IgG and MBLA).

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