EFFECT OF TRANSPLANTATION OF LYMPHOCYTES AND BONE MARROW CELLS ON IMMUNE RESPONSE DURING IMMUNIZATION OF MICE WITH LOW OR HIGH REACTIVITY TO THE ANTIGEN

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Mice of strains C57BL and CBA, with low and high reactivity respectively to immunization with sheep's red cells, received injections of syngeneic lymphocytes or bone marrow cells. Transplantation of syngeneic cells into C57BL mice increased the production of cells forming antibodies against sheep's red cells, but had a depressant effect in CBA mice. The different effects of transplantation of lymphocytes and bone marrow cells on the immune response, depending on the recipient's genotype, are discussed.
KEY WORDS: *immunogenetics; T-lymphocyte; B-lymphocyte; suppressor cells; phenotypic correlation.*

The writers showed previously [2, 3] that one cause of genetically determined differences in the immune response to immunization by sheep's red cells between CBA and C57BL strains of mice, with high and low levels of reactivity respectively, is the unequal intensity of cooperative interaction between B- and T-cells in these strains as a result of differences in the intensity of migration of these cells from the bone marrow and thymus. In other words, C57BL mice are characterized by a deficiency of cells of bone marrow or thymus origin, committed to sheep's red cells, which migrate from the corresponding organs; this also determines the lower ability of these mice to form antibody producers in response to immunization by this particular antigen than in CBA mice.

In this investigation the effect of transplantation of syngeneic lymphocytes and bone marrow cells on production of antibody forming cells (AFCs) was studied in C57BL and CBA mice immunized with sheep's red cells.

EXPERIMENTAL METHOD

CBA and C57BL mice aged 2-3 months were used. Cells obtained from bone marrow (1×10^7), lymph nodes (1×10^6 and 1×10^7), and thymus (2×10^7) were injected intravenously, either separately or together, into syngeneic recipients immunized intravenously with washed sheep's red cells (2×10^8). In the experiments of series I, the bone marrow, lymph node, and thymus cells were injected simultaneously with the sheep's red cells, whereas in series II the cells were transplanted either 24 h before or 24 h after immunization with sheep's red cells.

The number of AFCs in the spleen was counted by Jerne's method [10] on the 4th day after immunization if the lymphocytes and sheep's red cells were injected simultaneously, or on the 5th day after immunization if they were injected at different times.

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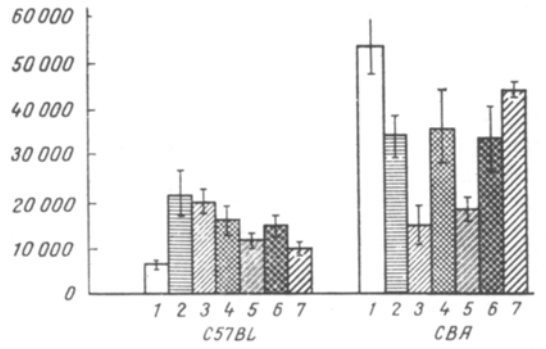

Fig. 1

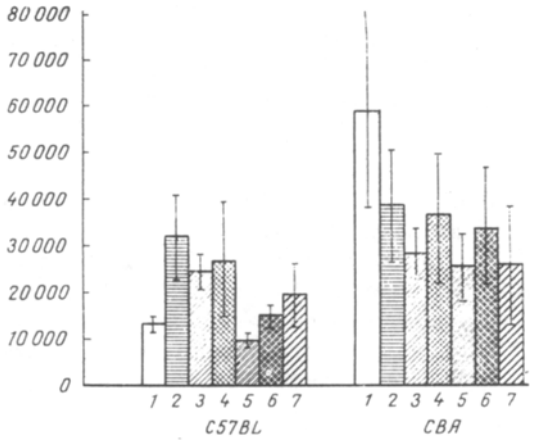

Fig. 2

Fig. 1. Number of AFCs in spleen of C57BL and CBA mice receiving different types of cells simultaneously with sheep's red cells: 1) injection of sheep's red cells alone; 2) the same + 10^7 bone marrow cells; 3) the same + 10^7 lymph node cells; 4) the same + 10^7 bone marrow cells and 10^7 lymph node cells; 5) the same + 10^6 lymph node cells; 6) the same + 10^7 bone marrow cells and 10^6 lymph node cells; 7) the same + 2×10^7 thymus cells. Here and in Fig. 2: ordinate, number of AFCs in spleen.

Fig. 2. Number of AFCs in spleen of C57BL and CBA mice receiving cells of different types before or after immunization with sheep's red cells: 1) injection of sheep's red cells alone; 2) the same, followed by bone marrow cells 24 h later; 3) the same, followed by lymph node cells 24 h later; 4) the same, followed by bone marrow and lymph node cells 24 h later; 5) bone marrow cells followed by sheep's red cells 24 h later; 6) lymph node cells followed by sheep's red cells 24 h later; 7) bone marrow and lymph node cells followed by sheep's red cells 24 h later.

The numerical results were subjected to statistical analysis [1] with calculation of the arithmetic mean (M), the standard error of the mean (m), and the 95% confidence limits ($P \leqslant 0.05$).

EXPERIMENTAL RESULTS

The results of the experiments in which bone marrow, lymph node, and thymus cells were transplanted simultaneously with immunization with sheep's red cells are illustrated in Fig. 1. During immunization of the C57BL mice, with low reactivity to sheep's red cells, 6400 AFCs were counted in their spleen. Combined or separate transplantation of syngeneic bone marrow and lymph node cells, and also of thymus cells into C57BL mice, immunized simultaneously with sheep's red cells, increased the immune response to that antigen several times over.

Immunization of CBA mice, with high reactivity to sheep's red cells, led to the production of 53,652 AFCs in their spleen. However, transplantation of 1×10^7 syngeneic bone marrow cells of this strain led to inhibition of the immune response. After injection of 1×10^7 lymph node cells, maximal suppression of the immune response was observed (14,882 AFCs were counted in the spleen). Much more marked inhibition of the immune response occurred after injection of a mixture of bone marrow and lymph node cells into the CBA mice. When 1×10^6 lymph node cells were transplanted, maximal suppression of the immune response again developed. The addition of 1×10^7 bone marrow cells to this dose of lymph node cells, as in the previous case, largely abolished the suppressive activity of the lymph node cells. Minimal suppression of the immune response in the CBA mice was observed after transplantation of thymus cells into them (44,710 AFCs).

Lymphocytes transplanted into CBA mice simultaneously with their immunization with sheep's red cells thus depressed their immune response to this antigen. The suppression was most marked when lymph node cells were injected and least marked when thymus cells were given. The addition of bone marrow cells to the inoculum of lymph node cells reduced their suppressive effect, even though bone marrow cells themselves also depress immunogenesis.

The results of experiments in which the lymphocytes were transplanted 24 h before or 24 h after immunization with sheep's red cells are given in Fig. 2. If bone marrow cells and lymphoid cells were transplanted separately or together into C57BL mice 24 h after immunization with sheep's red cells the immune response was intensified. If a mixture of bone marrow and lymph node cells was transplanted 24 h before immunization, a very small increase in the immune response was observed, whereas injection of bone marrow or lymph node cells alone had no effect on immunogenesis.

In the case of transplantation of cells into CBA mice 24 h after antigenic stimulation, suppression of the immune response was observed only if lymph node cells were given. Consequently, in CBA mice, belonging to a strain with high reactivity to sheep's red cells, transplantation of lymphocytes in most experiments led to suppression of the immune response. Maximal inhibition of the immune response in CBA mice occurred after transplantation of lymph node cells.

Depending on the genotype of the mice, transplantation of lymphocytes or bone marrow cells thus had different effects on the immune response induced by injection of sheep's red cells.

The enhancement of this response in C57BL mice can tentatively be linked with an increase in the number of B-cells committed to sheep's red cells in these animals, for the pool of these B-cells is considerably replenished after transplantation of bone marrow and lymph node cells. Unlike C57BL mice, CBA mice are not deficient in T- and B-cells committed to sheep's red cells. It is therefore perfectly logical to assume that injection of a further large number of "superfluous" cells will inhibit the development of the immune response, for the suppressive feedback mechanism can give a warning of such an excess. Investigations yielding evidence of the regulatory role of suppressor cells, which inhibit the immune response, have recently been published [4-7, 9]. Suppressor T-cells can regulate the activity of B-cells through a feedback mechanism. The presence of cells of macrophagal origin inhibiting the function of helper T-cells has been proved [8, 11, 12]. In the present experiments maximal suppression of the immune response in CBA mice was observed after transplantation of lymph node cells, suppression was less marked after injection of bone marrow cells, and it was minimal after transplantation of thymus cells. Consequently, cells inhibiting the immune response of normal CBA mice, of exogenous origin, may be present in different concentrations in different organs. In further research it would be interesting to study the origin and type of cells which alter the immune response of normal mice depending on their genotype.

LITERATURE CITED

1. I. P. Ashmarin and A. A. Vorob'ev, Statistical Method in Microbiological Research [in Russian], Leningrad (1962).
2. A. A. Batyrbekov, in: Collected Research Publications of Central Research Laboratories of Higher Medical Establishments of Uzbekistan [in Russian], Vol. 2, Tashkent and Samarkand (1974).
3. R. M. Khaitov and A. A. Batyrbekov, Byull. Éksperim. Biol. i Med., No. 2, 84 (1975).
4. A. C. Allison, A. M. Denman, and K. D. Barnes, Lancet, 2, 135 (1971).
5. P. G. Baker, P. W. Stashak, D. R. Amsbaugh, et al., J. Immunol., 105, 1581 (1970).
6. T. M. Chused, A. D. Steinberg, and L. M. Parker, J. Immunol., 111, 52 (1973).
7. W. Droege, Nature, 234, 549 (1971).
8. H. M. Folch et al., J. Immunol., 110, 835 (1973).
9. R. K. Gershon, R. Cohen, R. Hencin, et al., J. Immunol., 108, 586 (1972).
10. N. K. Jerne and A. A. Nordin, Science, 140, 405 (1963).
11. R. M. E. Paskhouse and R. W. Dutton, J. Immunol., 97, 663 (1966).
12. M. T. Scott, Cell. Immunol., 5, 459 (1972).