ABILITY OF CELLS OF THE REGENERATING SPLEEN TO EFFECT THE GRAFT VERSUS HOST REACTION

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The ability of cells of the regenerating (after single or twice repeated resection) and intact spleen of mice to induce the graft versus host reaction was studied by two methods. The regenerating spleen was shown to be less capable than the intact of bringing about this reaction.
KEY WORDS: *regenerating spleen; graft versus host reaction.*

The spleen is known to be able to regenerate after resection of a considerable part of its tissue [1, 2]. However, some functions of the regenerating spleen recover slowly. For instance, 1 month after resection, cells of the regenerating spleen form fewer antibodies in response to injection of thymus-independent Vi-antigen than cells of the control spleen [1]. Recovery of function of the regenerating spleen depends on the presence of the thymus, for after removal of the thymus the number of antibody-forming cells (AFCs) is reduced [2]. A similar process of a reduction in the number of plaques has been observed after subcutaneous autografting of pieces of spleen, evidently on account of their regeneration [4].

It was considered important to assess the function of the T-cells in the regenerating spleen. For this purpose the graft versus host reaction (GVHR), in which a leading role is played by the T-cells [5], was used.

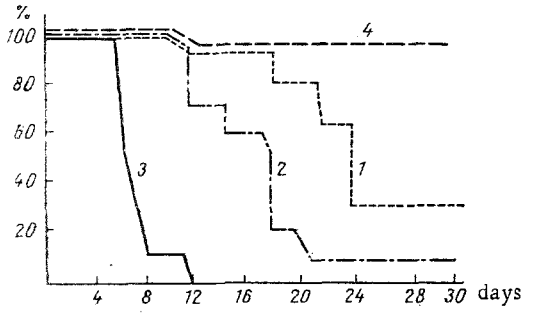

Fig. 1. Participation of regenerating spleen cells in GVHR: 1) period of regeneration 7 days; 2) 40 days; 3) intact spleen; 4) irradiation control. Abscissa, time after transplantation (in days). Ordinate, number of surviving recipients (in %).

EXPERIMENTAL METHOD

Experiments were carried out on 120 male CBA and (CBA × C57BL)/6 hybrid mice. In the experiments of group 1 the GVHR was investigated in irradiated recipients [7]. Two-thirds of the spleen was removed from CBA mice. A week or 40 days after the operation a cell suspension prepared from regenerating or intact spleens was injected in a dose of $3 \cdot 10^7$ cells into irradiated (600 R) hybrid recipients. The number of surviving mice in the experimental and control groups was compared over a period of 1 month.

In group 2 [6, 7] two-thirds of the spleen was resected in CBA mice. Ten days later, a further half of the regenerating spleen was removed from some of the mice recovering from the operation. One week after the first and after the second operation a suspension of

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TABLE 1. Number of AFCs in Spleens of Recipient Mice (M±m)

Group of experiments	Material injected into recipients	Mean number of AFCs per recipient's spleen
1st	—	14 893±12 630
	Intact spleen cells	4 720±850
	Cells of regenerating spleen (after 1 resection)	13 067±1 720
2nd	—	160 500±42 900
	Intact spleen cells	6 000±1 820
	Cells of regenerating spleen (after a 2nd resection)	52 160±7 260

cells from the regenerating or intact spleens of CBA mice was injected in a dose of $5 \cdot 10^7$ into unirradiated hybrid recipients. Seven days later they were immunized with sheep's red cells ($2 \cdot 10^9$). After 4 days the number of AFCs was determined in the spleen of the recipients by the local hemolysis in gel method [3]. The numerical results were subjected to statistical analysis.

EXPERIMENTAL RESULTS

As Fig. 1 shows, the dose of irradiation used was not lethal for the mice of this strain. Between 90 and 95% of the animals survived until the 15th day after irradiation.

After transplantation of spleen cells of a parental strain into a hybrid, death of the recipients takes place early and is due to acute homologous disease [7]. In fact, after transplantation of intact spleen cells into recipient mice, diarrhoea and emaciation were observed, followed by death of the animals in the course of 2 weeks (Fig. 1, 3). Recipients of regenerating spleen cells died much later (Fig. 1, 1, 2). Some animals survived for 1 month. Moreover, recipients receiving cells from the spleen after a longer period of regeneration (40 days; Fig. 1, 2) died sooner than the others. Consequently, the shorter the time of regeneration of the spleen after resection, the weaker the GVHR.

The results show that the function of the T-cells in the regenerating spleen was considerably depressed. The longer survival of the recipients could also be explained by a deficiency of T-cells in the regenerating organ. This hypothesis was confirmed by the results of the 2nd group of experiments. It will be clear from Table 1 that after injection of intact spleen cells of the parental strain into the hybrid the number of AFCs reacting to sheep's red cells was considerably reduced. This depression of the immune response was weaker after injection of regenerating spleen cells (13,067 ± 1720). Depression of antibody formation was particularly weakened after transplantation of spleen cells after a second resection of its tissue (Table 1).

A study of histological sections obtained during development of the GVHR by regenerating spleen cells on the 4th day after injection of sheep's red cells revealed a marked difference in the morphology of the structures responsible for antibody formation. For instance, after injection of regenerating spleen cells and treatment with antigen, multiple centers of proliferation developed in the Malpighian corpuscles of the spleen and the number of plasma cells increased. During the development of the GVHR by intact spleen cells and injection of sheep's red cells, only solitary small centers of proliferation and a few plasma cells were present.

It can be concluded from previous [2] and the present observations that the function not only of B-lymphocytes is reduced, as was demonstrated previously [1, 2], but also the immunologic activity of the T-lymphocytes, as shown by depression of the ability of regenerating spleen cells to induce the GVHR. This depression of functional activity was probably due to a decrease in the size of the T-cell population or to depression of the function of individual cells. Of course, the possibility that both these processes occur cannot be ruled out.

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