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FUNCTIONAL ACTIVITY OF NATURAL KILLER CELLS AND KILLER T CELLS IN MICE AFTER OVARIAN TRANSPLANTATION

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Prevention of the reaction of immunologic incompatibility during transplantation of foreign organs and tissues is a complex and still incompletely studied region of transplantology [1, 7]. A particularly urgent aspect is the study of the function and role of cytotoxic effector systems and, in particular, of natural killer (NK) cells and of killer T cells in transplantation.

The writers previously studied general and special aspects of the function of these lymphocyte subpopulations during transplantation of hematopoietic tissues [3]. In this paper we give data on the study of NK cells and killer T cells after allografting of the ovaries in mice.

EXPERIMENTAL METHOD

Mice of inbred lines CBA ($H-2^k$), C57BL/6 ($H-2^b$), and first-generation hybrids (CBA × C57BL/6)F_1, obtained from the "Stolbovaya" Laboratory Animals Nursery, Academy of Medical Sciences of the USSR, were used. Subcutaneous transplantation of CBA ovaries into C57BL/6 mice was carried out under aseptic conditions and hexobarbital anesthesia [8]. Some animals underwent a mock operation (without transplantation) under similar conditions. C57BL/6 mice of the same litter were used as the control group.

NK activity was determined in the membranotoxic test with ^{3}H-uridine [6]. Human erythromyeloid leukemia K-562 cells, maintained in culture [9], were used as target cells. Killer T cells were tested in the experimental system suggested by R. V. Petrov and co-workers [4]. Intact (CBA × C57BL/6)F_1 hybrid mice, after irradiation in a dose of 650 R, received transplanted lymph node cells ($1 \cdot 10^6$) from C57BL/6 mice: intact, subjected to a mock operation, or receiving an allograft of the ovaries (CBA). Lymph node cells were transplanted at different times (3, 7, 15, and 30 days) after the operation.

EXPERIMENTAL RESULTS

The experiments showed that in the early period after ovarian transplantation the membranotoxic activity of the recipients' NK cells was significantly (by 1.5-2 times) depressed compared with the control values. It can be tentatively suggested that suppression of NK activity at these times was due to stress, which is known to cause acute immunosuppres-

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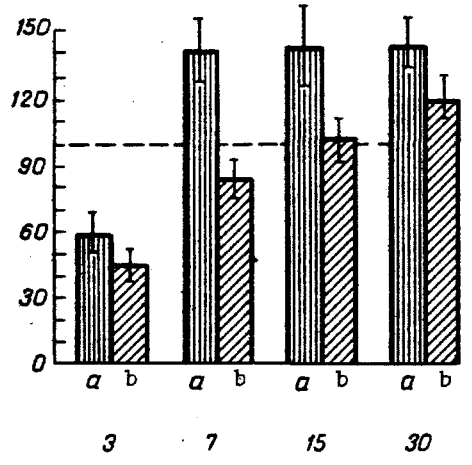

Fig. 1

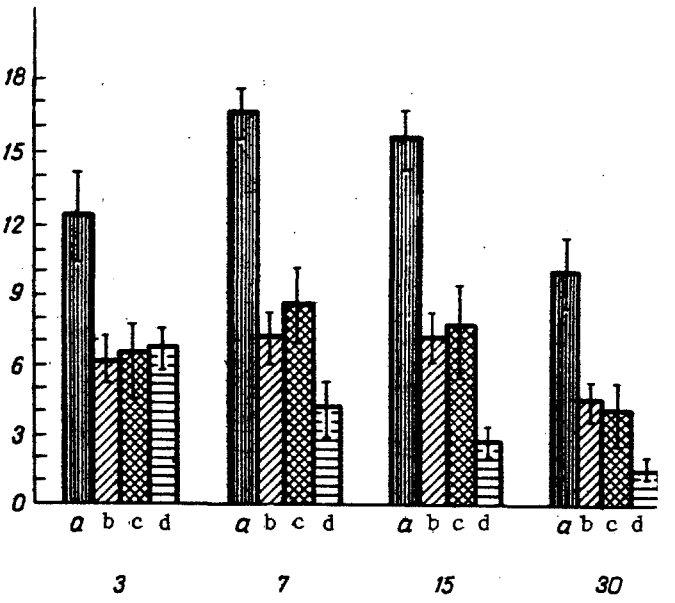

Fig. 2

Fig. 1. Membranotoxic activity of NK cells of C57BL/6 mice after transplantation of allogeneic ovaries from CBA mice. Ordinate, membranotoxicity (in per cent); abscissa — groups: a) transplantation of ovaries, b) mock operation, 3, 7, 15, 30) times of investigation (in days after transplantation); broken line — control.

Fig. 2. Activity of killer T cells of C57BL/6 mice after transplantation of allogeneic ovaries of CBA mice. Ordinate, number of endogenous CFU per spleen. Abscissa, days of investigation and groups: a) irradiation control, b) intact mice, c) mice undergoing mock operation, d) mice receiving graft.

sion in general [2] and suppression of NK cells in particular [5] in the body. Evidence to a certain degree in support of this view is given by the acute inhibition of NK cells which we found in the group of animals undergoing the mock operation.

At the later stages of the investigation (7th day) NK activity rose sharply (by 1.4 times) in receipients of the allogeneic ovaries, whereas the natural membranotoxicity of the lymphocytes of animals undergoing the mock operation was not yet restored, and did not reach the control levels (Fig. 1). Dissociation of NK activity in the recipients of the allogeneic ovaries and in those undergoing the mock operation discovered at these times undoubtedly indicates involvement (recruiting) of NK cells into the graft rejection reaction.

An elevated level of natural membranotoxicity of the recipients' lymphocytes also was recorded at subsequent times of the investigation. Incidentally, activity of NK cells at these same times in the group of animals undergoing a mock operation was completely restored and was virtually indistinguishable from the control level.

The study of the dynamics of changes in the killer T-cell system showed that in response to sublethal irradiation of (CBA × C57BL/6)F_1 mice on average 12.3 ± 1.7 endogenous colonies (CFU) were formed in their spleens. Transfer of lymph node cells (killer T cells) of intact C57BL/6 mice led to the formation of a much smaller number of CFU: the number of colonies was 6.1 ± 0.7 (degree of inhibition 50.5%).

Injection of lymphocytes from mice of the parental C57BL/6 genotype and undergoing a mock operation, on the 3rd day after the operation caused similar and comparable changes in the number of CFU, which in this case was 6.5 ± 1.3 (inhibition of colony formation by 47%).

A similar pattern in principle also was recorded after injection of lymphocytes from mice receiving the graft into the recipients (average number of CFU 6.8 ± 1.0, inhibition of colony formation by 45%). In other words, functional activity of killer T cells in the early period after transplantation, just as after a mock operation, remained substantially unchanged.

A rather different picture was observed on the 7th day after the operation. In the spleens of mice of the control group 16.7 ± 1.4 CFU accumulated. Transfer of lymphocytes from intact mice was accompanied by inhibition of proliferation of endogenous stem cells: inactivation of 56% of colonies (7.3 ± 1.3 CFU). Similar changes also were found after transfer of lymphocytes from recipients of killer T cells into animals undergoing a mock operation (8.7 ± 1.6): The degree of inhibition of colony formation was 48.0%.

At these times activation of killer T cells was recorded in mice receiving a graft. In the case of transfer of the lymphocytes of these mice into irradiated recipients, a high degree (74%) of inhibition of endogenous colony formation was observed.

Activation of cytotoxic killer T cells also was a characteristic feature of the later stages of the investigation.

It can thus be concluded from the experimental results described above that in response to transplantation of allogeneic ovaries marked activation of cytotoxic and membranotoxic effector mechanisms, which reflect the "intensity" [1] of transplantation immunity and can serve as a prognostic sign of graft rejection, takes place. An analysis is needed of the connection of these subpopulations of effector lymphocytes with other immunoregulatory classes of lymphocytes (suppressor T cells, countersuppressors) and cytokines of the immune system (interleukin-1, interleukin-2, etc.), and this will be a topic for our future research.

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