PRODUCTION OF ACTIVE IMMUNITY IN IRRADIATED ANIMALS BY ENTERAL VACCINATION

L. A. Nikolaeva

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Enteral vaccination of irradiated mice in accordance with a certain program causes the animals to develop immunity against typhoid bacilli equal in intensity to the immunity of unirradiated mice.

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Previous investigations have shown that enteral vaccination with heat-killed typhoid vaccine carried out on the 3rd day after irradiation increases the resistance of irradiated mice to infection with typhoid bacilli. These results are evidence of an immunologic response to administration of antigen in the irradiated organism.

The object of the present investigation was to study the dynamics of the immune response of the irradiated animal to enterally administered antigen.

EXPERIMENTAL METHOD

Experiments were carried out on noninbred albino mice weighing 18-20 g. The mice were irradiated on EKU-50 apparatus in a dose of 517 R($LD_{40-50/30}$). Dose rate 241 R/min. Corpuscular (heat-killed typhoid vaccine in a sessional dose of 5 billion bacterial cells), chemical (typhoid Vi-antigen obtained by the method of Webster and Landy as modified by V. V. Klyucheva, dose 1 mg), and living (Vi-containing strain Escherichia coli 5396/38, dose 2.5 billion bacterial cells) vaccines were used. Sessional doses of the vaccines were titrated in irradiated mice. All preparations were given in 0.2 ml physiological saline on the 3rd day after irradiation, on 3 successive days (1 cycle). At intervals of 7, 16, and 25 days after vaccination the mice were inoculated intraperitoneally with 1:4 dilutions of an 18-h living culture of typhoid bacilli strain Salmonella typhi ty_2 4446 (with agar). LD_{50} of the culture was calculated by Karber's method [1]. The experimental results were evaluated by the use of an index of resistance (IR; ratio between LD_{50} of the culture for the experimental animals and LD_{50} of the culture for intact animals). The natural resistance of the intact animals was taken as 1.

EXPERIMENTAL RESULTS

Irradiation in a dose of 517 R ($LD_{40-50/30}$) caused a sharp decrease in the natural resistance of the mice to infection with typhoid bacilli (Fig. 1). On the 12th day after irradiation the natural resistance was lowered by 15-20 times, but on the subsequent days it gradually recovered to almost the initial level of natural resistance of the intact animals.

After a single cycle of enteral vaccination carried out on the 3rd day after irradiation, the resistance of the irradiated mice to infection was significantly increased.

After vaccination with heat-killed typhoid vaccine the natural resistance of the irradiated animals was not depressed on the 12th day after irradiation (7th day after vaccination), but had returned to the original level of resistance of intact mice (IR = 1.0). By the 21st day after irradiation (16th day after vaccination) active immunity had developed in the vaccinated mice (IR = 15.6), its level corresponding to the intensity of immunity in unirradiated mice on the 7th day after vaccination. By the end of one month, the level of immunity was somewhat lower (IR = 9.3).

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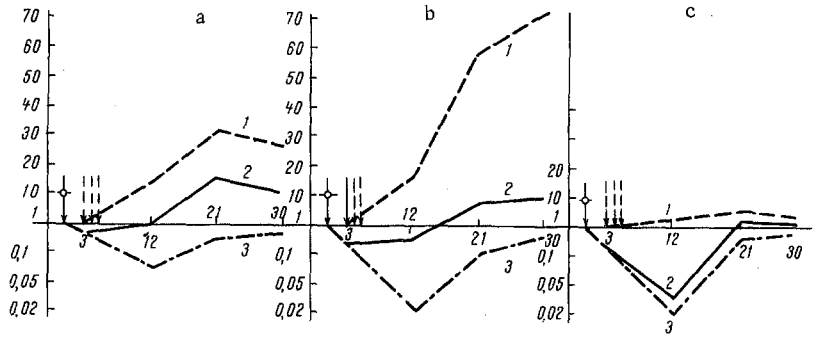

Fig. 1. Dynamics of resistance of mice to infection with thyphoid bacilli. Abscissa) interval (in days) between irradiation and infection; ordinate) index of resistance. 1) Unirradiated, vaccinated; 2) irradiated, vaccinated; 3) irradiated, unvaccinated mice. a) Heat-killed vaccine; b) Vi-antigen; c) living vaccine. Arrow with circle denotes irradiation; broken arrows denote vaccination.

TABLE 1. Resistance of Irradiated Mice (on 30th day after irradiation) Vaccinated Enterally (2 cycles) with Heat-Killed Typhoid Vaccine

Interval between vaccinations	Interval bet. 2nd cycle of vacc. and infec.	IR	
		irradiated	unirradiated
16 days (21)	7 days	27,6	24,3
7 days (12)	16 days	8,0	18,1
Unvaccinated		0,5	1,0

Note. Day after irradiation when 2nd cycle of vaccination began is shown in parentheses.

A similar picture was observed after vaccination with Vi-antigen. Active immunity was also found on the 21st day after irradiation (IR = 5.0), but it was lower in its intensity than after vaccination with heat-killed vaccine (15.6). In both cases the intensity of active immunity was indistinguishable from that in unirradiated animals on the 7th day after vaccination with the corresponding vaccines.

When enteral vaccination was carried out with a living culture of Escherichia coli, recovery of natural immunity took place sooner (by the 21st day after irradiation), but no active immunity developed. In all probability this was because the immunizing dose of vaccine was too small, as indicated by the low immune response in unirradiated animals. The use of large doses of living vaccine for immunization of irradiated animals is impossible because of the increased sensitivity of irradiated animals to microorganisms and their toxins.

Comparison of the curves reflecting resistance of irradiated, vaccinated and irradiated, unvaccinated mice to infection with typhoid bacilli thus revealed the stimulating action of enteral vaccination on resistance of the irradiated organism. A single cycle of enteral vaccination carried out on the 3rd day after irradiation not only led to earlier recovery of natural resistance, but also produced active immunity.

Comparison of the dynamics of the immune response to enterally administered antigen in irradiated and unirradiated mice revealed the similarity between them. However, the level of the immune response in the irradiated animals was much lower than in the unirradiated. In addition, the formation of active immunity in irradiated mice develops later, after the necessary time for recovery of the lowered natural resistance to the level of resistance of intact animals.

The time required for recovery of the lowered natural resistance of the irradiated animals differs depending on the nature of the vaccine. The fastest recovery was observed after enteral vaccination with heat-killed typhoid vaccine (12th day after irradiation), recovery was somewhat slower after vaccination with Vi-antigen (16th day), and it was slower still after vaccination with living vaccine (21st day after irradiation). In other words, depending on the type of vaccine, enteral vaccination accelerates the recovery of lowered natural resistance of irradiated mice by 1 (living vaccine), 2 (Vi-antigen), or 2.5 (heat-killed) vaccine) weeks.

It should be emphasized that on the 7th day after the lowered natural resistance had returned to normal, the irradiated mice had developed active immunity. Its intensity was indistinguishable from that of active immunity of unirradiated mice, also on the 7th day after vaccination with the corresponding vaccines.

In irradiated mice, therefore, the immune response to enteral administration of antigen is delayed by comparison with unirradiated animals. These results are in agreement with those obtained by other workers using different models or different ways of adminstration of the antigen, but also revealing delay of the immune response in irradiated animals [2, 3, 4, 6].

It can be concluded from these results that it is possible, in principle, to create active immunity by a single cycle of enteral vaccination of the irradiated animal on the 3rd day after irradiation.

Despite the fact that, as a result of one cycle of enteral vaccination, the irradiated animal develops active immunity, repeated vaccinations are necessary in order not only to maintain its level, but also to raise it.

Experiments were carried out to study the effectiveness of two cycles of enteral vaccination of irradiated mice with intervals of 7 and 16 days between cycles, so that the second cycle of vaccination took place on the 12th or 21st day after irradiation. As Table 1 shows, the best effect of enteral vaccination was obtained by the use of a double cycle of vaccination with an interval of 16 days between cycles. When this scheme of vaccination was used, the intensity of immunity developed by the irradiated animal (IR = 27.6) was indistinguishable from that of immunity in the unirradiated mice. In irradiated unvaccinated mice at this time (30 days) the level of natural immunity was approximately the same as normally (IR = 0.5). After two cycles of vaccination, but at an interval of 7 days, i.e., when the second cycle was given 12 days after irradiation, active immunity also developed by the 30th day after irradiation, but its intensity was lower (IR = 8.0).

It can thus be concluded from these results that under certain conditions it is possible for active immunity to be produced in an irradiated animal by enteral vaccination, and the intensity, of that immunity is at least equal to that observed in the unirradiated animal.

LITERATURE CITED

1. I. P. Ashmarin and A. A. Vorob'ev, Statistical Methods in Microbiological Research [in Russian], Leningrad (1962).
2. D. R. Kaulen, Med. Radiol., No. 6, 51 (1956).
3. D. R. Kaulen, Med. Radiol., No. 2, 65 (1957).
4. H. J. Kohn, J. Immunol., 66, 525 (1951).
5. M. S. Silverman and P. H. Chin, J. Immunol., 66, 525 (1951).
6. M. S. Silverman and P. H. Chin, J. Immunol., 77, 266 (1956).