EFFECT OF ANTIBIOTIC THERAPY ON THE COURSE OF HOMOLOGOUS SICKNESS IN F_1 MOUSE HYBRIDS

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Prolonged administration of oxytetracycline and streptomycin to (CBA×C57BL/6)/F_1 mouse hybrids into which the spleen cells of C57BL/6 mice had been transplanted increased the life span of the animals. These results are interpreted as proof of the role of autoinfection in the pathogenesis of homologous sickness.

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A previous investigation [2] showed that in homologous sickness developing in (CBA×C57BL/6)/F_1 hybrids after injection of parent spleen cells an autoinfection regularly appears and can be detected in most mice in the late stage of the desease. The question arises whether this autoinfection is merely the result of the diseases, an accompanying factor, or whether it plays a definite pathogenetic role in the development of this syndrome.

The object of the present investigation was to test the hypothesis that if the observed autoinfection is one cause of death of the animals from transplantation sickness, intensive antibiotic therapy should prevent or depress the development of this disease.

EXPERIMENTAL METHOD

Homologous sickness was produced by injecting living parent spleen cells of adult C57BL/6 mice in a dose of $110 \cdot 10^6$-$120 \cdot 10^6$ into the retroorbital venous sinus of unirradiated (CBA×C57BL/6)/F_1 hybrids aged 6-8 weeks and weighing 16-18 g. The suspension of cells was obtained by mincing spleen tissue in a glass homogenizer with the addition of Hanks' solution, followed by passage several times through a Kapron filter. Eosin in a dilution of 1:2000 was used to count the dead cells. The suspension was kept on ice and injected into the mice 30-60 min ater preparation. Before injection, heparin was added to the suspension in a dose of 5-10 units per mouse. Aseptic precautions were observed throughout the investigation.

After transplantation of spleen cells, the experimental mice received streptomycin and oxytetracycline with the food for 100 days. Altogether 6 experiments were carried out, in which 75 experimental and 60 control mice were used. Hybrids receiving injections of parent spleen cells only acted as controls.

For the first 5 days after the beginning of treatment the mice received 50,000 units streptomycin and 10,000 units oxytetracycline daily per animal. Later the antibiotics were given every other day in half this dose. The administration of antibiotics started on the 2nd, 5th, 10th, and 25th days after transplantation of spleen cells.

EXPERIMENTAL RESULTS

The experimental results are given in Table 1. In the control hybrids not receiving antibiotics, signs of the disease began to appear from the 13th-16th day after transplantation (decrease in body weight, wasting, untidiness of the hair, diarrhea). The mean survival period of the mice was 27-36 days and the mortality varied from 90 to 100%.

Administration of the antibiotics caused a marked improvement in the course of the homologous sickness. Antibiotic therapy was particularly effective if started on the 2nd day after transplantation of spleen cells (Table 1, experiment No. 1): neither development of disease nor death of the experimental animals

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TABLE 1. Effect of Antibiotic Therapy on the Course of Homologous Sickness in F_1 Mouse Hybrids

Experiment No.	Group of mice	No. of mice in group	Day of starting treatment after injection of spleen cells	No. of mice dying within 100 days	Mean life span of mice (in days)	P
1	Experimental	10	2	–	> 100	< 0.01
	Control	10	–	9	27.0 ± 1.9	–
2	Experimental	15	5	8	67.3 ± 3.3	< 0.01
	Control	10	–	10	28.7 ± 1.6	–
3	Experimental	10	5	8	49.0 ± 7.1	< 0.01
	Control	10	–	10	26.2 ± 2.6	–
4	Experimental	10	5	1	92	< 0.01
	Control	10	–	7	35.4 ± 5.7	–
5	Experimental	10	10	9	49.3 ± 3.1	< 0.01
	Control	10	–	10	27.4 ± 2.2	–
6	Experimental	20	20	19	49.0 ± 1.9	< 0.01
	Control	10	–	10	36.9 ± 3.1	–

was observed in the course of 100 days (period of observation), whereas in the control group 90% of the mice died within 30 days after transplanation. In this particular experiment administration of the antibiotics was stopped after 100 days. Eight days after discontinuing the antibiotics, these mice showed signs of transplantation sickness, from which 4 of the animals died 5-7 days after the onset of the disease (the remaining mice were sacrificed). At necropsy signs of atrophy of the lymphoid organs were found in all the dying animals and in 3 of those which were sacrified. A similar worsening of the course of the homologous sickness was observed after discontinuation of antibiotic therapy in 7 of the 8 mice in experiment No. 2 and in 3 of the 9 mice in experiment No. 4.

If antibiotic therapy began on the 5th-10th day after transplantation (experiments No. 2, 3, 4, 5), in all cases a significant increase in the life span of the experimental mice was observed compared with the controls ($P < 0.01$). Antibiotic therapy had a marked effect even if started at the height of the homologous sickness, when some animals had already died (experiment No. 6): the condition of the mice improved, their diarrhea disappeared, and their period of survival was significantly prolonged by comparison with the control ($P < 0.01$). The 15 mice whose condition again deteriorated a short time after the beginning of antibiotic therapy, despite the continuation of treatment, were sacrificed. Usually animals were killed when it became clear that they would die very shortly. Seedings were made from the heart, liver, spleen, and mesenteric lymph glands of the sacrificed mice in serum broth and on blood agar.

The results of the bacteriological investigation showed that bacteria of the intestinal group were cultured from one or several organs of 13 of the 15 animals, in most cases resistant to the antibiotics used for treatment. The isolated bacteria were identified by the methods used previously [2].

Hence, massive antibiotic therapy can depress or prevent the development of homologous sickness produced in hybrids by injection of parent spleen cells.

It can be concluded from these results that the autoinfection developing in F_1 hybrids with homologous sickness is an important factor in the pathogenesis of this syndrome. In some cases, it is evidently the immediate cause of death of the animals, while in others it can be responsible for the development of marked pathological changes [1, 3] in various organs, particularly the intestine, which can aggravate the course of the homologous sickness.

LITERATURE CITED

1. V. G. Molotkov and E. A. Fedosov, in: Proceedings of the 26th Scientific Conference of Smolensk Medical Institute on Problems in Noninfectious Immunology [in Russian], Smolensk (1968), p. 21.
2. A. S. Shevelev, E. A. Fedosov, and M. M. Kirvel', in: Proceedings of the 26th Scientific Conference of Smolensk Medical Institute on Problems in Noninfectious Immunology [in Russian], Smolensk (1968), p. 50.
3. E. A. Cornelius, C. Martinez, E. J. Yumis, et al., Transplantation, 6, 33 (1968).