OPPOSITE ACTION OF QUINGAMINE ON SKIN ALLOGRAFT REJECTION AND ANTIBODY-FORMING CELL LEVEL IN MICE

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Great attention is currently being paid to the study of the immunomodulating properties of drugs used in clinical practice in the form of long-term courses, for example, quingamine* [10]. Besides its use for the treatment and prevention of malaria, this drug is also used in the treatment of lupus erythematosus, rheumatoid arthritis, mononucleosis, psoriasis, and hepatitis [10]. In addition to its ability to interfere with metabolic processes in the cell [5, 8] and to exert an anti-inflammatory action [12], quingamine also possesses immunodepressive activity [2, 6, 12]. The character and importance of the immunomodulating action of quingamine during the treatment of malaria and of autoimmune diseases, however, have not been adequately studied [12].

This paper describes the results of a study of the effect of quingamine in various doses on the rejection time of a skin allograft and on the level of antibody-forming cells (AFC).

EXPERIMENTAL METHOD

To determine the action of quingamine on B-cell immunity, male (CBA × C57BL/6 Jax) hybrid mice weighing 18-20 g, obtained from the Stolbovaya nursery, Academy of Medical Sciences of the USSR, were used. The effect of the preparation was determined by counting the number of AFC in the spleens of the mice, using the method [11] in the modification [3]. Sheep's red blood cells (SRBC) were injected intravenously in a single dose of $5 \cdot 10^8$ cells. The day of injection of SRBC was taken as day 0. Quingamine was injected intraperitoneally into the mice before their immunization with SRBC (days −) once or 4 times, and also after their immunization with SRBC (days +) once. The results were subjected to statistical analysis by Student's test [1]. To study the effect of quingamine on rejection of skin allografts, male CBA mice were used. C57BL/6 Jax mice served as the donors. Skin allografts were transplanted by the method in [7]. The results were read as the time of complete rejection of the skin graft, taking the day of transplantation of the graft as day 0. Quingamine was injected intraperitoneally into CBA mice before skin grafting in doses of 20, 30, 40, and 50 mg/kg 4 times, or 25 mg/kg twice. The results were subjected to statistical analysis by Wilcoxon's test [1].

EXPERIMENTAL RESULTS

The experimental results given in Table 1 show that four preliminary injections of quingamine (on days −4, −3, −2, and −1) in doses of 50 and 40 mg/kg lengthened the survival of the skin allograft on average by 5 days ($p < 0.05$). Injection of quingamine in a smaller dose (30 mg/kg) by a similar scheme lengthened the survival of the skin graft by 3.2 days ($p < 0.05$). Reducing the dose of the drug to 20 mg/kg, but administering it by the same scheme or as two injections at an interval of 24 h between them had no effect on the survival time of the skin graft ($p > 0.05$).

The next series of experiments showed that four preliminary injections of quingamine in a dose of 50 mg/kg, with intervals of 7 days between injections, caused significant lengthening of the survival of the skin graft (Table 1). The results of the present experiments must be compared with those obtained previously by other workers. For instance, it was shown that

*Quingamine is a proprietary preparation of chloroquine diphosphate — Translator.

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TABLE 1. Effect of Preliminary Injection of Quingamine on Rejection of Skin Allografts in CBA Mice (C57BL/6 donors)

Expt. No.	Group No.	Dose, mg/kg	Number of injections	Interval between injections, days	Interval between injection and transplantation, days	Time of injection, days	$M \pm m$	U test
1	1	Control	—	—	—	12, 12, 12, 15, 15, 15, 12	13,9±1,6	$p>0,05$
	2	25	2	1	0	12, 12, 12, 12, 14, 16, 16	13,4±1,9	
2	1	20	4	1	1	11, 11, 9, 10, 12, 8, 8, 11	9,9±1,5	$p_{1-5}>0,05$
	2	30	4	1	1	15, 16, 13, 11, 13, 16, 16, 16	14,3±1,9	$p_{2-5}=0,005$
	3	40	4	1	1	18, 18, 18, 11, 19, 13	16,1±3,3	$p_{3-5}=0,005$
	4	50	4	1	1	16, 17, 16, 17, 16, 16, 15	16,1±0,6	$p_{4-5}=0,01$
	5	Control	—	—	—	11, 11, 10, 11, 11, 11, 13	11,1±0,9	—
3	1	50	4	7	5	16, 16, 16, 16, 16, 16, 17, 18	16,3±0,7	$p<0,01$
	2	Control	—	—	—	10, 10, 10, 10, 13, 13, 14, 14, 15	12,1±2,1	

TABLE 2. Effect of Preliminary Course and Single Injection of Quingamine on Formation of AFC to SRBC, Injected on Day 0

Group No.	Time of injection of quingamine, days	Dose, mg/kg	Number of animals	Geometric mean number of AFC per spleen	Significance	Effect
1	—4 —3 —2 —1	20 (4 times)	10	416 330 (5,619±0,051)	$t=2,34$ $p_{1-3}<0,05$	Stimulation
2	—4 —3 —2 —1	40 (4 times)	10	430 232 (5,634±0,028)	$t=3,8$ $p_{2-3}<0,01$	Stimulation
3	—	Control	8	304 334 (5,483±0,028)	—	—
4	—2	40	5	301 360 (5,479±0,115)	$t=0,2$ $p_{4-10}>0,05$	—
5	—2	20	5	430 934 (5,634±0,033)	$t=2,87$ $p_{5-10}<0,05$	Stimulation
6	—1	40	10	356 233 (5,552±0,06)	$t=1,24$ $p_{6-10}>0,05$	—
7	—1	20	9	388 897 (5,490±0,048)	$t=0,5$ $p_{7-10}>0,05$	—
8	0	40	10	260 844 (5,416±0,03)	$p_{8-10}>0,05$ $t=0,5$	—
9	0	20	10	171 205 282 904	$t=2,5$ $p_{9-10}<0,05$	Inhibition
10	Control	—	10	(5,452±0,054)	—	—

Legend. Here and in Table 3: in one version of the experiments injection of smaller doses (10 and 5 mg/kg) had no effect on the AFC level; values of M ± m given in parentheses.

TABLE 3. Effect of Quingamine on Formation of AFC to SRBC Injected on Day 0 (single injection)

Group No.	Time of injection of quingamine, days	Dose, mg/kg	Number of animals	Geometric mean number of AFC per spleen	Significance	Effect
1	+1	20	8	354 624 (5,549±0,02)	$p>0,05$ $t=1,5=1,2$	—
2	+1	40	7	310 198 (5,491±0,02)	$p>0,05$	—
3	+2	20	8	219 114 (5,341±0,083)	$p>0,05$	—
4	+2	40	7	245 763 (5,391±0,170)	$p>0,05$	—
5	Control	—	8	304 344 (5,483±0,028)	—	—

treatment of animals with quingamine (in a daily dose of 40 mg/kg), starting from the day of skin grafting and continuing on the following days, until total rejection of the skin allograft, had no effect on the length of its survival [9]. Thus the times and dose of the compound injected play an important role in determining the immunodepressive action of quingamine on rejection of skin grafts.

The effect of quingamine on the AFC level was studied by the use of different doses of the drug and different times of its injection. After four preliminary injections of quingamine in doses of 20 and 40 mg/kg significant stimulation of AFC formation was found. When a single injection of the drug was given 2 days before immunization of the animals (day −2) quingamine, in a dose of 20 mg/kg, likewise stimulated AFC formation. When a double dose (40 mg/kg) of the drug was given at these same times, it had no effect on the AFC level (Table 2). Injection of quingamine on day 0 in a dose of 20 mg/kg caused a significant fall of the AFC level. However, the same dose of the drug (20 mg/kg) had no action on the number of AFC when it was injected after immunization with SRBC (Table 3). Thus in different modifications of the experiments, when different doses of quingamine were used, it was found to have an opposite action.

In the discussion of these results the most probable hypothesis is evidently that which postulates that stimulation of the AFC level is evidently unconnected with the action of quingamine itself, but is a manifestation of the activity of its metabolites.

It is also logical to suggest that quingamine metabolites affect different T cell populations. In particular, the results of the present experiments do not rule out their effect on T suppressor and T helper cells. It is easy to imagine in such a case that the thresholds of sensitivity of different cell populations may differ.

The results evidently also admit the possibility of an opposite action not only of quingamine itself and of its metabolites, but also of similar relations between the metabolites of this drug. The essential fact here is that quangamine accumulates in high concentrations in the liver and spleen [4]; 50% of the drug, moreover, is excreted unchanged by the kidneys, and 25% of it is excreted in the form of metabolites [5, 8]. Another important fact is the presence of the unchanged drug in the body for 7-10 days after its last injection [4].

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